Author:
Wainwright, Benito

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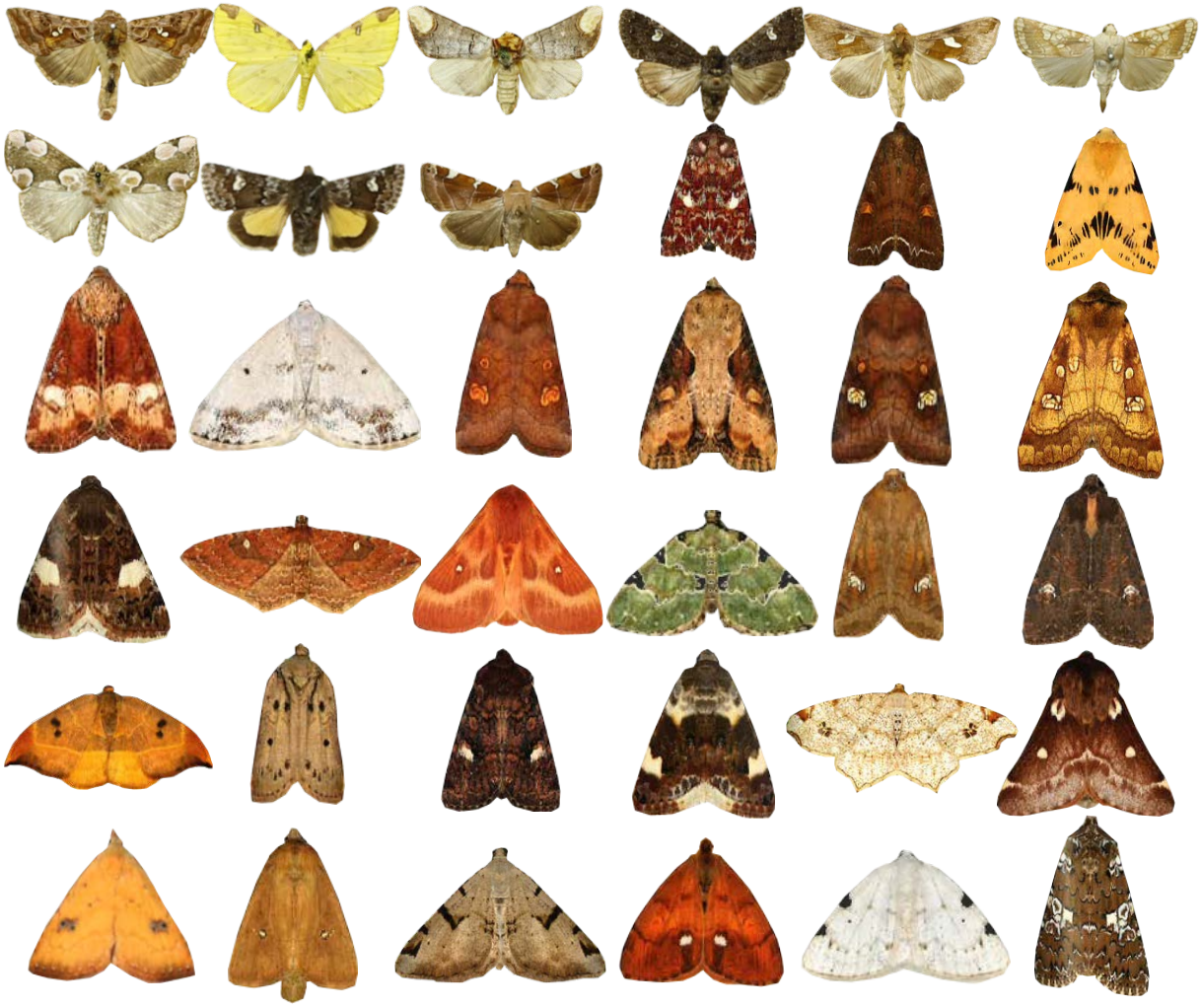
MSc project by Benito Wainwright



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Abstract

For camouflaged prey animals which benefit from remaining undetected by predators, increased conspicuousness due to bilaterally symmetrical colouration is costly. The ubiquity of symmetrical body patterns in nature is assumed to be due to tight genetic and developmental constraints. Given these evolutionary restrictions, we investigated how animals might have evolved to optimise their surface colouration in order to reduce the predation cost of bilaterally symmetrical colouration. One way of doing this is by placing discrete, high contrast markings away from the axis of symmetry (or midline). Artificial camouflaged prey with symmetry placed at different distances from the axis were used in both laboratory visual search tasks with humans and field survival experiments with wild avian predators. By measuring response time (Experiment 1) and predation rate (Experiment 2), I showed that for targets that were symmetrical only near their midline, detectability was the same as for targets that were symmetrical all over their surface. Prey were significantly less conspicuous when symmetry was placed outside of this 'critical zone'. To see whether living animals have evolved as predicted from the trends suggested by these experiments, the saliency of features at different distances from the midline of the cryptically coloured forewings from 36 Lepidopteran species was measured. Measures of both absolute salience and relative to wing area demonstrated that salience is indeed greatest away from the 'critical zone' near the axis of symmetry. My results suggest similarities in the mechanisms of bilateral symmetry detection by mammalian and avian predators. They also suggest that evolution has resulted in pattern elements being positioned in such a way that the detectability cost imposed by symmetry is reduced.

Keywords: bilateral symmetry; camouflage; Lepidoptera; predation; salience; visual search

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Author's Declaration

I declare that the work in this dissertation was carried out in accordance with the requirements of the University's *Regulations and Code of Practice for Research Degree Programmes* and that it has not been submitted for any other academic award. Except where indicated by specific reference in the text, the work is the candidate's own work. Work done in collaboration with, or with the assistance of, others, is indicated as such. Any views expressed in the dissertation are those of the author.

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1. A literature review on the link between animal camouflage and bilateral symmetry

1.1 Background

The threat of predation has resulted in the evolution of a myriad of animal defensive strategies (Ruxton *et al.,* 2004; Ruxton *et al.,* 2018). Prey with visually oriented predators have evolved colours and patterns to ,distract, deter and deceive their enemies (Stevens, 2016). Perhaps the most widespread of these is camouflage, whereby an animal's colours and patterns decrease the probability of detection whilst the animal remains within the visual range of a predator (Stevens & Merilaita 2009a, 2011; Nokelainen & Stevens, 2016; Merilaita *et al.,* 2017). Initially noted as an excellent example of the workings of evolution by natural selection (Darwin, 1859; Wallace 1877, 1889; Poulton 1890), the mechanisms by which animal camouflage works were not laid out in any great detail until artist Abbott Thayer (1896) and zoologist Hugh Cott (1940) published their influential work linking biology, art and the military. Somewhat later on, Endler (1981) attempted to categorise types of colouration in terms of how they might influence the perception of the predator. However, it has only been over the past couple of decades that these ideas have started to be empirically tested. Now, with the establishment of more rigorous experimental methods, many of the theories proposed at the end of the 19th and first half of the 20th century are beginning to gain quantitative support (Stevens & Merilaita, 2009a; Ruxton *et al.,* 2018).

The study of the mechanisms by which animal camouflage strategies have evolved to deceive the predator's eye, both historically and to the present day, has drawn ideas from Gestalt psychology (Metzger, 1936; Behrens, 1980, 2002; Cuthill & Troscianko, 2009; Troscianko *et al.,* 2009; Osorio & Cuthill, 2015). According to this early and influential school of perceptual psychology, there are cues, such as proximity or symmetry of surface pattern elements, and continuity of edges, that determine how relevant objects are segregated from their background and, more generally, how perceptual scenes are organised (Troscianko *et al.,* 2009; Osorio & Cuthill, 2015). These fundamental principles of perception generate selection pressures, influencing the evolution of aspects of the body surface coloration, shape and behaviour which reduce or distort the effect of these cues and thereby decrease the likelihood of detection and/or recognition (Cuthill *et al.,* 2007; Troscianko *et al.,* 2009; Osorio & Cuthill, 2015; Merilaita *et al.,* 2017).

The shape of the body outline is a cue which not only is important in recognition, but potentially makes an organism more salient against its background. This is because, even with excellent matching of colour and texture to the average background, unless the background is featureless or very homogeneous there are likely to be discontinuities in colour and pattern phase at the body's edge. In order to reduce such effects, many animals have evolved disruptive colouration: markings

that break up the body outline or other distinctive features, an idea first outlined by Thayer (1909). Cott (1940) formalised Thayer's observations of disruptive colouration into three main principles: differential blending, maximum disruptive contrast, and coincident disruptive colouration. Differential blending is the close match of some (but not, in Cott's formulation, all) colour patches to colours in the background (Stevens *et al.,* 2006; Stevens & Merilaita, 2009b). Maximum disruptive contrast is a strong internal contrast between colour patches on the animal, such that some colour patches do not match the background (Merilaita, 1998; Schaefer & Stobbe, 2006; Stevens *et al.,* 2006). Coincident disruptive colouration is continuity of contrasting colour patches across adjacent body parts (e.g. limbs held next to the body) such that the distinctive shape of those body parts is broken up (Cott, 1940; Cuthill & Szekely, 2009). It was not until much later that these hypotheses and observations were empirically tested, and in large part supported, demonstrating disruptive colouration to be distinct from a simple resemblance to the colour and visual texture of its background (Merilaita, 1998; Cuthill *et al.,* 2005). Disruptive colouration exploits different mechanisms of the predator's perceptual processing, both low- and high-level, in order to avoid detection (Espinosa & Cuthill 2014) and it is a strategy now believed to be taxonomically widespread (Ruxton *et al.,* 2018). The methodologies that enabled the investigation of how animals may optimise the crypsis of their body outline can be applied to another major Gestalt cue: the bilateral symmetry of surface colours and patterns.

1.2 An introduction to symmetry in appearance

1.2.1 Early ideas on symmetry and camouflage

Most metazoans have a plane of (usually bilateral) symmetry (Finnerty *et al.,* 2004; Palmer, 1996). Cott (1940) studied closely how the characteristics of an animal's outline facilitate recognition, but symmetry was not something which he focused on in any detail. He was nevertheless aware that it was a characteristic which facilitated visual recognition, despite the lack of experimental evidence beyond human perceptual psychology. One of Darwin's contemporaries however, Alfred Russell Wallace, wrote at some length of how having a plane of symmetry increases the risk of predation (Wallace, 1867, 1879, 1891). He even theorised that many species of stick insect (order Phasmotodea) might have evolved to optimise their camouflage by exposing their legs asymmetrically when at rest (a behavioural response) in order to mitigate this cost (Wallace, 1891, pp. 47).

The idea of symmetrical objects being more efficiently segmented from cluttered backgrounds than asymmetrical objects has been known in human experimental psychology for some time (Barlow & Reeves, 1979), being one of the features that Gestalt psychologists identified (Metzger, 1936; Osorio

& Cuthill, 2015). It has been described as a fundamental image feature (Rock, 1983), and discriminability of random dot displays improves with increasing degree of symmetry for human participants (Barlow & Reeves, 1979). To apply these principles of human perception to the context of animal colouration, one must first identify whether non-human animals can indeed perceive bilateral symmetry. Surprisingly few studies have tested this explicitly, but the data that have been published thus far indicate that this ability has evolved in multiple taxa.

1.2.2 Symmetry perception in non-human animals

Osorio (1996) used a model to demonstrate that symmetry can be computed by mechanisms which are present in the early visual pathways. The perception of symmetry has been studied in particular depth in the context of plant-pollinator interactions whereby honeybees (*Apis mellifera*) have been shown to have innate preferences for symmetrical flowers (Giurfa *et al.,* 1996). Once symmetry is detected, it can be learnt and generalised, allowing bees to respond to novel stimuli and to discriminate between radially and bilaterally symmetrical patterns (Horridge, 1996). However, a problem with using honeybees for such studies is that it is extremely difficult to control their visual experience before the experiment, due to the near-impossibility of keeping completely indoor communities. Bumblebees (*Bombus terrestris*), on the other hand, can be kept completely naïve and, when similar studies are conducted with these organisms, innate preferences for bilateral symmetry are still observed (Rodríguez *et al.,* 2004).

Similar psychophysical experiments have been conducted in many of the major vertebrate taxa. In fish, this perceptual ability appears to vary between species: for example, bamboo sharks (*Chiloscyllium griseum*) demonstrate exceptional spontaneous symmetry preference and discrimination, whereas distantly related Malawi cichlids show no innate preference and little discrimination between symmetrical and asymmetrical patterns (Schluessel *et al.,* 2014). In birds, pigeons (*Columba livia*), starlings (*Sturnus vulgaris*) and chickens (*Gallus gallus*) have been the primary study organisms where symmetry detection, discrimination and generalisation have been explicitly shown (Delius & Habers, 1978; Swaddle & Pruett-Jones, 2001; Swaddle *et al.,* 2008; Mascalzoni *et al.,* 2015), but this ability has also been inferred in studies on other avian species (e.g. Møller, 1993). The degree to which the perceptual capabilities of humans generalises to other mammals is sparsely researched, but early work on apes and dolphins (Rensch, 1958; Paukner *et al.,* 2017; Von Fersen *et al.,* 1992) has suggested that they, too, are able to recognise symmetrical patterns. To date, there has been no similar published work demonstrating symmetry perception in reptiles or amphibians. Psychophysics provides fundamental information on the relevance of

symmetry perception to the sensory ecology of the taxon in question. From here, one can begin to make experimental manipulations in order to investigate its putative role in defensive coloration.

1.3 The cost of symmetrical colouration

Cuthill *et al.* (2006a) conducted the first field experiments investigating the role of pattern symmetry in camouflage. Artificial moth-like targets with completely symmetrical and asymmetrical wing patterns were pinned on to oak trees and left exposed to wild bird predation under natural conditions. Checks at regular intervals showed that the predation rates of moths with symmetrical patterns were significantly greater, suggesting that having symmetrical body patterns imposes a significant intrinsic cost on animals that benefit from remaining undetected. Further work, using the same experimental design in the field, showed that this cost applied equally to background matching and disruptive patterns (Cuthill *et al.,* 2006b; figure 1.1(a),(b)). This suggests that the cost of bilateral symmetry reduces the efficacy of both of these defence strategies, and is a widespread hindrance to the camouflage of many animal species. The role of symmetry in animal camouflage therefore has both ecological and evolutionary relevance given the seemingly paradoxical diversity of animals which are bilaterally symmetrical in their surface patterning.

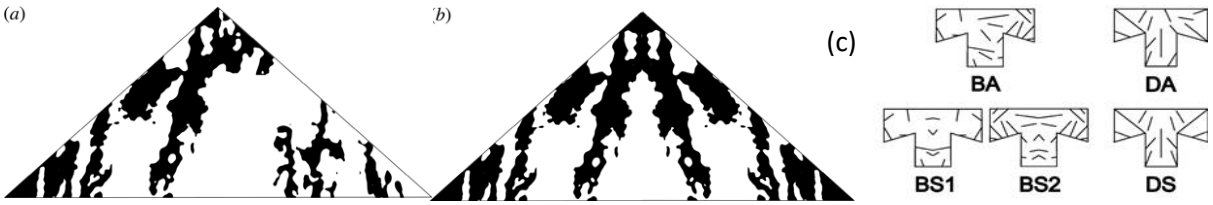


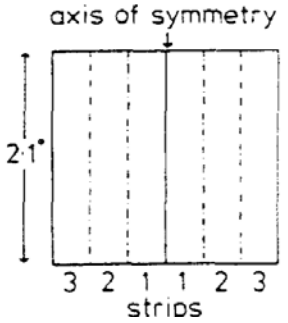
Figure 1.1. (*a*) Examples of the 2 (*a*) asymmetrical and (*b*) symmetric treatment groups used by Cuthill *et al.* 2006a in their wild predation experiments. (*c*) The targets used by Merilaita & Lind (2006) in their captive great tit experiments, showing the background matching and disruptive asymmetrical patterns (BA and DA), and the background matching symmetric (BS1 and BS2) and disruptive symmetric (DS) patterns.

Since the initial quantitative clarification of the inherent cost of asymmetrical patterns, it remains relatively unexplored whether some symmetrical patterns are less salient than others (Merilaita *et al.* 2017). Nevertheless, a laboratory predation experiment with captive great tits (*Parus major*) showed that the relative cost of symmetry varied between patterns (Merilaita & Lind 2006, figure 1.1 (c)). A limitation recognised by the authors was that when creating the symmetrical background-matching targets, pattern elements that were not present in the background were inadvertently synthesised, whereby straight lines were converted into chevrons (see also Discussion in Cuthill & Troscianko, 2009). These may have significantly increased the detectability of the symmetrical patterns due to the reduced match between the target and the background. Another more interesting explanation, not totally separate from the latter, is that patterns closer to the axis of symmetry are especially important in symmetry detection. It is this principle and its applications that I shall investigate in more detail.

1.4 Symmetry perception in human experimental psychology

1.4.1 Random-dot studies

The symmetry of the positioning of surface pattern elements on the body is a significant aspect of animal cryptic colouration which must be taken into consideration (Corballis & Roldan, 1975; Swaddle, 1999). The effect of the pattern midline had been known for several years to be of disproportionate importance in the saliency of symmetrical shapes. Human perception experiments have shown that not only are small violations in symmetry less easily detected with increased distance from the midline, but also that the detection time is shorter than detecting slight changes in repeated patterns (Bruce & Morgan, 1975). This highlights the effectiveness of symmetry as an easily extracted Gestalt cue, particularly in visually oriented animals, presumably because points near the symmetry axis are closer to the point of fixation (Barlow & Reeves, 1979; Locher & Nodine, 1989). Follow-up experiments using random dot displays with different degrees of mirror symmetry have further demonstrated the plasticity of symmetry perception when viewed with changing orientation and when the axis is not central in the field of vision (Barlow & Reeves, 1979). A "six-slice experiment" was performed by these authors, where each figure was divided into six and the symmetry between corresponding pairs was altered (figure 1.2). Discriminability significantly increased when symmetry was located at the pair of strips closest to the symmetry axis, but dots on the pair of outer strips were more easily discriminated than dots in the middle pair. The outer pair often determined the outer contour of the entire target, so it was probably this that made these areas overall more salient to the human subjects. It is, however, possible that this finding is an artefact of the particular stimuli (random-dot targets) used; in starlings, individuals are able to discriminate between differences in symmetry in natural-looking stimuli, but not with artificial random-dot targets (Swaddle & Pruett-Jones, 2001; Swaddle & Ruff, 2004; Swaddle *et al.,* 2008), thus emphasising the need to use more naturalistic stimuli when testing the perceptual capabilities of non-human animals.



strips	d′ BR	HB
1	4·6	3·6
2	0·97	0·76
3	1·8	2·7

Figure 1.2. An illustration of the design used by Barlow & Reeves (1979) (left). Symmetrical random dots were placed in one of the 3 pairs of strips, leaving the rest asymmetrical. The results (right) show that discriminability (d') was greatest when symmetrical pairs of dots were adjacent to the axis of symmetry. Symmetrical pairs of dots on the outer strip were the next most effective.

The early work described above has since been confirmed (e.g. Wenderoth, 1994; Tyler *et al.,* 1995). Barlow & Reeves (1979) previously suggested that the presence of bilateral symmetry allows an image to be encoded "economically", as only half of a symmetrical pattern needs to be computed for complete image perception. Jenkins (1982) investigated whether visual systems gave a disproportionally greater weight to points nearer the axis of symmetry of random dot displays. The results revealed the presence of a symmetry threshold close to the axis which must be exceeded in order for symmetry to be perceived. All other symmetry information beyond this point was deemed to be relatively redundant in its contribution to symmetry detection, and this was shown to be independent of stimulus size. However, it remains unclear whether these findings were specific to the dot displays used or whether they could be generalised to visual search for real objects in real scenes.

1.4.2 Studies with biologically relevant stimuli

It was not until several years later that a number of new studies were published, the results of which had more relevance to biological systems. Gurnsey *et al.* (1998) attempted to model the visual mechanisms by which symmetry detection facilitates object segmentation under more natural viewing conditions where the axis is not necessarily centred in the visual field: their data suggested that symmetry does not in fact contribute significantly to image segmentation in isolation, and that it only plays an important role once the object has been fixated. The saliency of symmetry is therefore not likely to be constant across all retinal locations, and its perception relies upon higher level visual processing than previously thought. Further work was carried out to demonstrate the importance of spatial filters in mirror symmetry integration (Rainville & Kingdom, 2000), particularly near the symmetry axis. Although the precise neurophysiological pathways underpinning how symmetry is encoded are not the primary concern in this literature review, they nevertheless provide a model for the perceptual capacity of animals. This model will ultimately determine the strength of the selection pressure on aspects of body patterning, leading to the evolution of features that deceive the visual system of the enemy.

Despite these endeavours to visualise how symmetry is interpreted by living systems, organisms nevertheless display a greater variety of complex shapes compared with the simple random dot patterns in the experiments previously described. Wilson & Wilkinson (2001) applied sums of radial frequency, quantified deviations from complete circularity (Wilkinson *et al.* 1998), to generate more complex shapes. This provided a precise method to quantify deviations from bilateral symmetry of more biologically relevant stimuli. It was found that the neural mechanisms underpinning the perception of these shapes differ from those used during random dot symmetry detection,

indicating the potential irrelevance of the latter stimuli to symmetry detection in natural scenes. These results provide a way for the symmetry of more relevant shapes to be experimentally tested. Evans *et al.* (1999) used simple image processing to create a human visual search task where participants were shown high resolution images of crabs and insects to investigate the effect of orientation of the axis in symmetry detection. Published studies of this kind are rare but they provide a benchmark for further investigation of the efficacy of symmetry relative to the spatially contiguous effect of the symmetry axis in a biological context.

1.5 Artificial neural networks and emergent properties of sensory systems

The incorporation of artificial neural networks into the study of animal colouration and perception is still relatively unexplored (Merilaita, 2007), but may prove highly informative in providing detailed models of the mechanisms by which patterns deceive predator perception. In an investigation looking at symmetry preference in mate choice, Enqvist & Arak (1994) and Johnstone (1994) incorporated neural networks to mimic a simple sensory system with the task of discriminating between objects of different sizes/lengths that were, on average, bilaterally symmetrical but which varied around a mean asymmetry of zero (see section 1.9). It was discovered that symmetry preference was simply a by-product (i.e. an emergent property) of how such a network learns categories: a symmetrical pattern was the mean of all other patterns presented, and thus evoked the strongest response even if it was not part of the training set. It would be of great interest to apply these techniques and ideas to more realistic representations of the sensory systems of different animal taxa, in order to see whether concealing symmetrical features affects these networks in similar ways.

1.6 Asymmetrical exceptions in vertebrates

For a trait to evolve by natural selection, the fitness benefits must outweigh the fitness costs of having the trait. Given that most animals are indeed symmetrical, this seems to be a trait which is either of high benefit itself, or the costs of deviating from it are high; for example, if there are tight genetic and developmental constraints. In spite of this, flatfish such as plaice, sole, turbot and halibut have managed to circumvent this constraint by expressing genes in pathways that determine levels of thyroid hormone which controls embryonic organ lateralization during larval metamorphosis (Tagawa & Aritaki, 2005; Schreiber, 2013). This is when the asymmetrical flatfish body plan is formed, making them the most asymmetrical of all vertebrates, with even their eyes positioned asymmetrically, allowing them to perceive downwelling light fully whilst resting on sandy

sea beds. Therefore, the evolution of asymmetrical body plans is achievable, but probably only under strong selective pressures. The camouflage of flatfish is so effective that it has become a selection pressure in itself, resulting in the evolution of interspecific mimicry by the Atlantic longarm octopus (*Macrotritipus defilippi*), which imitates the behaviour and morphology of the peacock flounder (*Bothus lunatus*) (Hanlon *et al.,* 2010).

While an asymmetrical body plan does have anatomical, mobility, and sensory costs associated with it, it is nevertheless not clear why asymmetrical colouration and patterning, superimposed on an underlying symmetrical body, are not more abundant in the animal kingdom. In addition to their asymmetrical anatomy, flatfish also demonstrate dynamic camouflage, giving them the flexibility to optimise their colouration to different backgrounds in a heterogeneous environment (Ramachandran *et al.* 1996). Further studies have found that plaice (*Pleuronectes platessa*) are able to alter the expression of spots and blotches actively on their body surface independently of one another, thus allowing optimisation of crypsis to specific backgrounds (Kelman *et al.,* 2006). Light and dark markings are roughly symmetrically distributed about the midline when the animal is viewed from above which, given the asymmetrical anatomy of these animals, suggests that there are no developmental constraints. Plaice also have markings which are not under dynamic control and these are largely distributed asymmetrically. A noteworthy observation was that the majority of the spots on the body surface were far less concentrated near the symmetry axis, rather being more prominent near the sides of the body. Unfortunately, no subsequent work has attempted to quantify this.

Other than those on flatfish, studies investigating asymmetry in body colour evolution in other vertebrate groups are next to non-existent, except in relation to the small random deviations, known as fluctuating asymmetry, in a normally bilaterally symmetrical trait (see section 1.9). When it comes to directional asymmetry, where large deviations from symmetry are the norm, Wallace (1889) noted that “bilateral symmetry of coloration is very frequently lost among domesticated animals, it almost universally prevails in a state of nature”. Osorio (1994) made the observation that the plumage of the wryneck, *Jynx torquilla*, a species of woodpecker, shows high degrees of asymmetry but there has been no rigorous experimental work on directional asymmetry in this species, or on any other bird for that matter!

1.7 Symmetry and camouflage in cephalopods

The taxon in which rapid dynamic camouflage is by far most impressive and widespread is the class Cephalopoda, a group of marine molluscs that includes squid, octopus and cuttlefish (Hanlon *et al.,* 2018). The latter in particular provide extraordinary examples of sophisticated, high-speed visual

control of camouflage patterns. Cuttlefish are able to change their appearance in less than a second through the fine neural control of chromatophore organs on the skin surface (Hanlon & Messenger, 2018). Studies of these colour and pattern changes under laboratory conditions facilitate our understanding of the Darwinian fitness benefits of different camouflage designs. Hanlon (2007) began to classify animal patterning into being either Uniform, Mottle or Disruptive and suggested that these may provide the scaffolding which will favour the evolution of cephalopod camouflage against a range of backgrounds, an idea which has been widely tested in cephalopods (Allen *et al.,* 2009; Barbosa *et al.,* 2008; Barbosa *et al..* 2008; Buresch *et al.,* 2011; Chiao *et al.,* 2009; Chiao *et al.,* 2005; Chlao *et al.,* 2007; Kelman *et al.,* 2007; Maethger *et al.,* 2007; Shohet *et al.,* 2007; Shohet *et al.,* 2006; Zylinski *et al.,* 2012; Zylinski *et al.,* 2016; Zylinski *et al.,* 2009).

Given the apparent plasticity of these animals when it comes to changing their appearance, they may provide ideal model organisms for the study of symmetry and camouflage (Langridge, 2006). It has been shown that cuttlefish are able to discriminate visually between potential threats and select the most appropriate defensive display accordingly (Langridge *et al.,* 2007). However, this has only been studied in great detail in the context of warning or "deimatic displays" whereby a high contrast signal is displayed to potential attackers (Umbers *et al.,* 2015). Nevertheless, it does suggest the possibility that these animals may be able to actively change their surface patterning depending on the predation risk (Adamo *et al.,* 2006; Langridge *et al.,* 2007).

The common cuttlefish, *Sepia officinalis*, is able to dynamically control the degree of bilateral symmetry in its surface colouration (Langridge 2006). The previously described genetic and developmental mechanisms constraining asymmetry evolution therefore appear not to be of relevance in these animals, making them prime model organisms for experimental manipulation. Langridge (2006) conducted a behavioural analysis of cryptic and deimatic displays by cuttlefish in order to investigate whether surface patterning is indeed more asymmetrical when an individual is camouflaged rather than displaying to a predator. In the case of the latter, theory suggested that display efficacy should increase as body patterning becomes more symmetrical (Forsman & Merilaita, 1999). The results were in fact surprising. In the 10 juvenile cuttlefish studied, the degree of bilateral symmetry was significantly greater for cryptic body patterns whereas the deimatic display was often expressed asymmetrically. This outcome seems to contradict the initial hypotheses of the study, as has been empirically demonstrated in laboratory and field conditions using models of bird and human vision (e.g. Barlow & Reeves, 1979; Cuthill *et al.,* 2006a,b; Merilaita & Lind, 2006). However, in Langridge's experiments, the predators (small bass) approached the cuttlefish from one side; it is on that side that the cuttlefish produced the spot, suddenly, and only when the predator was right upon it. This is consistent with a deimatic display and, given the lateral approach, a

bilaterally symmetrical deimatic display would provide no advantage. Other authors, based on the prevalence of symmetrical camouflage patterns in cuttlefish and octopus, have also argued that body pattern symmetry might somehow be advantageous, either for camouflage (although the precise mechanism is unknown) or as a display of fitness (Allen *et al.,* 2010) (see section 1.9). The difference in the relative energy investment for producing an asymmetrical rather than a symmetrical pattern in these animals is negligible. However, what their work actually suggests is that, for these animals on these backgrounds, asymmetry confers no significant benefit, not that symmetrical patterns provide better camouflage.

There are other explanations, taking account of the physiology and evolutionary ecology of cephalopod dynamic colouration, which might explain these seemingly anomalous results. As colour change is controlled by vision, there could be a mechanistic constraint whereby, if the information received by the two eyes is the same, as would be the case when against a cryptic background, then bilaterally symmetrical surface patterning might be an emergent default property of the colour change mechanism. However, the experimental designs of this study and previous ones (e.g. Hanlon & Messenger, 1988) state that this is unlikely. A more satisfactory explanation could be based upon field observations which suggest that teleost predators very rarely detect cryptic cuttlefish despite them being symmetrical (Hanlon & Messenger, 1988). Furthermore, when cuttlefish are resting on a seabed, for predators coming from different directions the orientation of the axis of symmetry will constantly differ in direction when viewed from above. This explains why even humans have difficulty detecting camouflaged cuttlefish, as it is well established that symmetry is more efficiently detected when the axis is vertical in the visual field (e.g. Barlow & Reeves, 1979; Corballis & Roldan, 1975; Wenderoth, 1994; Evans *et al.,* 2000; figure 1.3). The relative cost of bilateral symmetry is therefore likely to be higher for a prey item resting vertically (given that predators will generally be upright when approaching) than for one where the predator may approach from several directions.

The position of symmetrical features in relation to the axis of symmetry was not investigated by Langridge (2006) or Allen *et al*. (2010), but this might have provided an insight into whether symmetry is posing a constraint on the camouflage of these cuttlefish. The unpredictability of the orientation of the midline might therefore explain why cephalopod predators such as teleost fish could have a somewhat diluted degree of symmetry perception. This requires further psychophysical investigation and indeed, other authors have argued that rotationally invariant symmetry detection systems should be more advanced in animals which frequently encounter symmetrical stimuli at unpredictable orientations (Hollard & Delius, 1982). It is also worth noting that for a receiver viewing

a cuttlefish unilaterally, overall body symmetry does not matter, particularly in the case of the deimatic displays. Rather, one would expect selection for asymmetry of individual markings instead.

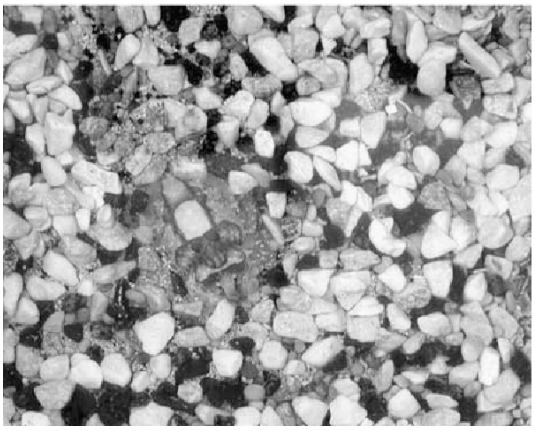


Figure 1.3. *Sepia officinalis* displaying an effective concealing pattern despite its bilateral symmetry (Langridge 2006)

Future studies require the incorporation of the dynamic patterning of cephalopod colouration with the perceptual capabilities of cephalopod predators. Because of the unpredictability of axis orientation, it is possible that the selective pressure for symmetry perception is somewhat diluted in predators of these marine invertebrates. The link between symmetry and camouflage in marine cephalopods is therefore more complex than previously envisaged.

1.8 Opposing ideas

There have, however, been theoretical arguments suggesting that symmetry may in fact be beneficial to camouflage. Troscianko *et al.* (2009) proposed that symmetry perception depletes a perceiver's cognitive resources. Figure-ground organisation, the mechanism by which a two-dimensional shape is converted into a three-dimensional one by the visual system of the perceiver, relies heavily on the computation of the symmetrical contours of the animal. This has been demonstrated in domesticated chicks, where recognition of three-dimensional form was facilitated in symmetrical objects (Mascalzoni *et al.,* 2015). The time taken for the object to be resolved by the receiver may provide the time necessary for the prey item to escape. This is a controversial possibility which lacks evidence, given that the static nature of the targets used by Cuthill *et al.* (2006) and Merilaita & Lind (2006) meant that it was not possible for them to "escape" upon detection. But ultimately, any time taken on the analysis of shape by the perceiver is surely still worse for the prey than not being detected in the first place! There is evidence of behavioural contour modification by a twig masquerading spider (Zhang *et al.* 2015), suggesting that this is an adaptive strategy that would enhance camouflage but, overall, this is an area which deserves more attention.

Bilaterally symmetrical organisms also have enhanced pattern regularity, as Cott (1940) himself observed. Experiments on blue tits in the laboratory have since shown that pattern regularity significantly increases detectability and therefore impairs camouflage (Dimitrova & Merilaita, 2012). Furthermore, experiments on poultry chicks show that bilateral symmetry facilitates figure-ground organisation (Mascalzoni *et al.* 2015) and would thereby reduce the time a predator would spend in recognising a prey as distinct from its background. Again, a more detailed knowledge of the neuropsychological mechanisms that take place during object detection and recognition, and their costs, is required before estimates of the relative Darwinian fitness payoffs can be made.

1.9 Fluctuating asymmetries and sexual selection

The opposing force acting against the evolution of body pattern asymmetry which has received the most substantial attention in the literature is sexual selection. Fluctuating asymmetries are small, random deviations from perfect symmetry in a bilaterally symmetrical trait where the mean is 0 and the distribution of traits is approximately normal (Palmer & Strobeck, 1986). Empirical work both in the field and in the laboratory has investigated the possibility that fluctuating asymmetry might be an honest measure of fitness when choosing between mates. It has therefore been claimed it is a powerful tool for investigating "good genes" models of sexual selection (Møller & Pomiankowski, 1993).

For example, the symmetry of secondary sexual ornaments has been analysed in various bird species from museum collections (Møller & Höglund, 1991). In this study, characteristics of sexual signalling were shown to have significantly higher degrees of symmetry than other morphological features, but a subsequent study did not replicate the findings (Balmford *et al.,* 1993). Follow-up studies on diverse species have since contributed to a wealth of literature in this area. The role of symmetry during signalling, not just in mate choice but also between flowering plants and their insect pollinators (Møller & Eriksson, 1994, 1995), is therefore an area of controversy and overall the link between symmetry and attractiveness is weak. For example, in Lepidoptera, intersexual comparisons of the symmetry of the speckled wood butterfly (*Pararge aegeria*) have shown variation in males to be directional rather than fluctuating (Windig & Nylin, 1999). Studies such as these provide explanations as to why the relative importance of the role of symmetry in sexual selection may vary between taxa.

There has been less literature focussing specifically on the link between sexual selection and the fluctuating asymmetry of body colouration as opposed to the size of morphological features. One of the most cited cases is a study conducted by Swaddle & Cuthill (1994a) in which a choice chamber

experiment showed that captive female zebra finches prefer males with symmetrically coloured leg bands over asymmetrically banded ones. However, there is very sparse literature investigating natural surface colouration, most probably because of the difficulty of manipulating colour patterns of live animals, especially without changing anatomical symmetry. Swaddle & Cuthill (1994b) manipulated the black-and-white barred chest plumage of male zebra finches and found a preference for symmetrical patterns, but their manipulation was not referenced to natural variation in the pattern and so, as with their leg band experiment, has doubtful ecological validity. Both experiments do, however, show that there is a preference for symmetrical patterns in this species. Badyaev *et al.* (2001) separated components of avian carotenoid-based sexual ornamentation, pigment and patch area asymmetry being one of them, to study how selection acts upon independent components which cumulatively give rise to the evolution of composite sexual traits. They also investigated how the extent of selection upon these individual components varied between populations. Asymmetries were assessed using digital imaging and it was found that selection for higher fecundity and viability was positively correlated with both pigment and patch area symmetry (figure 1.4).

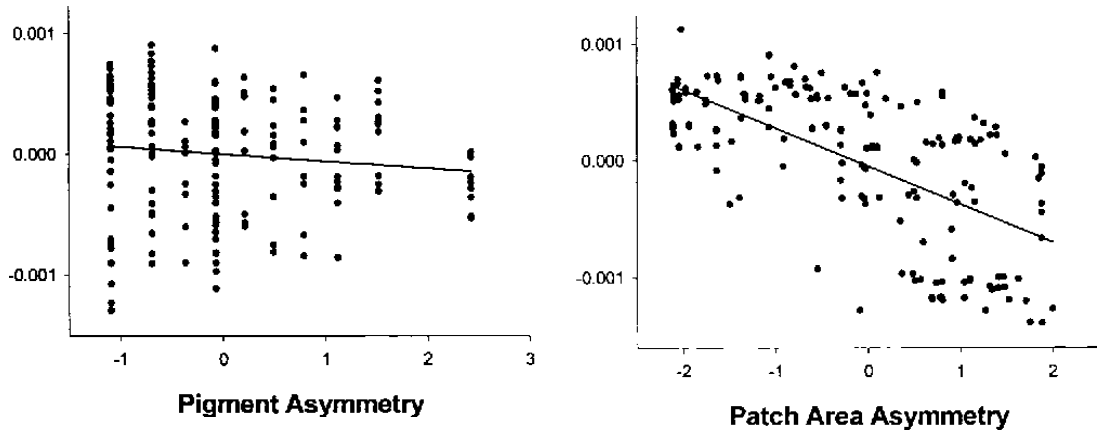


Figure 1.4. Partial regression plots from Badyaev *et al.* (2001) illustrating how pigment and patch area asymmetry in house finch breast ornamentation affects male net fitness (defined as breeding season fecundity and survival post-breeding season).

Fluctuating asymmetry in animal colouration therefore has the potential to signal fitness information, and a more detailed analysis will help to identify components of symmetry which improve saliency. The study by Badyaev *et al.* used the house finch (*Carpodacus mexicanus*) as a model species, but it has not yet been established whether the results generalise to other taxa or even other bird species. Work on the butterfly *Bicyclus anynana* suggested that female choice does not depend on eyespot symmetry (Breuker & Brakefield, 2002), but it is known that eyespots in this species serve other purposes, notably deflecting attack to non-vital body regions (Prudic *et al.,* 2015). As yet there has been no research to investigate even the optimum positioning of secondary

sexually signalling traits, let alone with respect to the axis of symmetry. Once such work is undertaken, a multivariate approach could be applied to test whether the trade-off between camouflage and sexual selection is real.

1.10 Aposematism and symmetry

It is not only to members of the same species that animals may wish to signal. Aposematic colouration has evolved in many taxa to signal distastefulness and unprofitability to predators (Guilford, 1990; Mappes *et al.,* 2005; Rojas *et al.,* 2015). Research into whether the efficacy of these signals improves with increased levels of symmetry was undertaken by exposing artificial prey to domestic chick predation (Forsman & Merilaita, 1999). The results suggest that predation should select for symmetrical warning displays as these facilitate detection and learning. Subsequent studies on laboratory chicks identified asymmetry detection thresholds beyond which the efficacy of conspicuous colour patterns is less effective (Forsman & Herrstrom, 2002). Unexpectedly, these results were not supported by survival analysis conducted in the field (Stevens *et al.,* 2009). This could be due to the nature of the targets used (e.g. the artificial prey used by Forsman & Merilaita were unpalatable whilst those used by Stevens *et al.* were not) and to the fact that the symmetry detection thresholds of wild avian predators are likely to be higher in the field because the background is more complex (Dimitrova & Merilaita, 2010; Xiao & Cuthill, 2016). However, it could be that other features such as size or colour have more important roles in neophobia or the generalisation of warning signals (which is what Stevens *et al.* 2009 were effectively studying), as opposed to the speed of aversion learning (which is what Forsman and colleagues investigated). These are factors which would merit further investigation.

It should be noted that camouflage and aposematism are unlikely to be opposing selective forces within the same species. With the exception of distance-dependent effects (Barnett *et al.,* 2017) and cases where a conspicuous hindwing may be concealed by a cryptic forewing in Lepidoptera (e.g. Forsman & Merilaita 2003), one would expect cryptic and warning colouration not to be used simultaneously. Animals may get around this by displaying multimodal sensory cues which convey different information to different receivers. However, on the whole, cryptic colouration should evolve alongside other features which also reduce the risk of detection. Therefore, asymmetry should be selected for in cryptically coloured species and symmetry in aposematic species; image analyses suggest that this could be the case (see section 1.12).

1.11 Genetic and developmental constraints in arthropods

Arthropods such as insects probably have even tighter constraints against surface asymmetry due to the mechanisms by which pigment is produced. Insects often incorporate pigment into their exoskeleton via sclerotization (Hopkins & Kramer, 1992) but, in the case of Lepidoptera, specialised cells in the epidermis can produce body pigment and pigment precursors (Wittkopp & Beldade, 2008). These pigments are deposited at the late pupal stage by specialised scale-building cells in the cuticle (Nijhout, 1980) and reaction-diffusion models have more recently been used to understand how features such as eyespots develop (Dilao & Sainhas, 2004). In vertebrates, body pigments are distributed via the migration of specialist cell types, which provides more developmental plasticity (Hoekstra, 2006).

A lot of developmental work has been conducted on the African nymphalid butterfly, *Bicyclus anynana*, which possesses prominent eyespots on its forewings. These, normally circular, eyespots have been artificially selected to be expressed ellipsoidally in order to work out trait correlations that affect wing shape (Monteiro *et al.,* 1997). The resulting phenotypes were subsequently analysed morphometrically to demonstrate that additive genetic variance for eyespot shape exists. Wing shape and colour patterns were shown to be developmentally coupled, suggesting that the decoupling of these characters may have a significant developmental cost because of this lack of independence between traits. Symmetry of wing shape has obvious locomotor benefits, meaning that genes influencing these traits will be favoured by natural selection, which in turn will favour the transmission of linked genes. This will lead to biases in the direction of evolutionary adaptation, leading to some phenotypes being more likely to evolve than others.

This view of evolution, that adaptations consists of few loci of large effect, is reminiscent of Sewell Wright's early theoretical work (1932) in which mathematical models revolve around the assumption that many loci have pleiotropic effects. As a result of this, a benefit in one phenotype may be overruled by potentially fatal consequences in others (Cheverud, 1984). These ideas are relevant to the evolution of asymmetrical patterning by suggesting that it would require a strong selective force in order to uncouple the relationship between symmetrical features and body development, which could be why asymmetrical patterning is relatively rare in arthropods in nature.

It is only a recent discovery that surface colours and patterns in Lepidoptera are controlled by a discrete set of genes, namely the *Wnt* complex. The *WntA* gene, specifically, has been identified as necessary for the induction of symmetry systems, a conserved set of pattern elements that are thought to be responsible for the development of most butterfly colours and markings (Martin & Reed, 2014; Mazo-Vargas *et al.,* 2017). Therefore, mutations in single developmental genes can have

cascading effects on the morphological complexity and diversity within a clade, perhaps hinting further at the pleiotropic interactions constraining the plasticity of surface patterning.

A noteworthy exception - an insect with asymmetrical colour patterns - is *Tithrone roseipennis*, a tropical mantis. In this species, a green wing conceals a red wing when at rest (Barabás & Hancock, 1999). When disturbed, the red forewing is revealed, presumably eliciting a startle response in the attacker, which decreases the risk of predation. Epistatic interactions between forewing colouration and wing folding are thought to have facilitated the evolution of these unusual phenotypes at the molecular level, although this is yet to be investigated explicitly. The more we understand about the underlying molecular and developmental pathways by which bilateral symmetry arises in the class Insecta, the more likely we are to have a fine-grained understanding of the factors constraining the evolution of asymmetry. *T. roseipennis* may provide a valuable model for evo-devo researchers to work with in the future.

1.12 A review of methodologies

A wide range of methodologies have been applied to test causal, developmental, functional, and evolutionary hypotheses concerning symmetry in animal colouration. However, no studies have applied all these dissimilar techniques to the same system.

1.12.1 Molecular approaches

An integrative approach would, for example, enable the quantification of selection on relevant genetic loci in the field and this has started to be done on genes such as *Mc1R*, involved in fur camouflage in mammals (Steiner *et al.,* 2007). With new genomic technologies becoming increasingly accurate and affordable, using genes as fundamental units of selection in order to follow the evolution of complex quantitative traits such as body pattern symmetry will undoubtedly become easier in the future.

1.12.2 Laboratory and field artificial target studies

The use of artificial targets in the analyses of predation rates under natural conditions has provided valuable insights into the field of animal visual defence strategies (e.g. Cuthill *et al.,* 2005; Mappes *et al.,* 2014). When field studies are combined with lab studies under controlled conditions (e.g. Merilaita & Lind, 2006) and previously described work from human perceptual psychology (e.g. Barlow & Reeves, 1979; Gurnsey *et al.,* 1998; Rainville & Kingdom, 1999), we can be more confident in making the generalisation that features such as asymmetrical body patterning affect the saliency of prey in the same way for both mammalian and avian visual systems. However, to date no human

visual search task has demonstrated the cost of symmetrical colouration on camouflage using natural-looking stimuli or against natural backgrounds. Nevertheless, bird and human experiments investigating other properties of camouflage patterning using biologically relevant targets show strong concordance with each other (e.g. Cuthill & Szekely, 2009; Stevens *et al.,* 2013; Xiao & Cuthill, 2016; Michalis *et al.,* 2017).

1.12.3 Image analysis

Digital photography of museum specimens provides a way of quantifying various aspects of an animal's colour and patterning, and is a technique that is slowly gaining popularity (Stevens *et al.,* 2007), particularly when used in conjunction with image analysis software (Troscianko & Stevens, 2015). One such example involved intra-individual comparisons of the symmetry of several species of the order Lepidoptera (Forsman & Merilaita, 2003). The aim was similar to that of the study by Langridge (2006), which compared the cryptic and deimatic patterns of cuttlefish. By using image analysis, Forsman & Merilaita (2003) investigated the relative symmetry of the cryptic forewings and signalling hindwings of the chosen study species. It was found that relative asymmetry did tend to be greater in the cryptic traits of the forewings compared with the aposematic signalling features of the hindwings (figure 1.5), but for the most part these differences were not statistically significant. The authors explained these results in the context of tight genetic and developmental constraints. However, the studies of dynamic colouration in the cephalopods indicate that symmetrical camouflage patterns may be produced even when this constraint is removed.

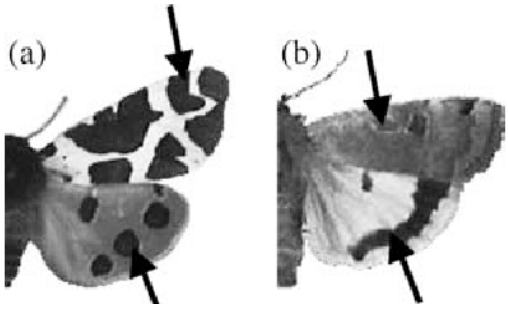


Figure 1.5. Illustrations of 2 moth species used by Forsman & Merilaita (2003) ((*a*) *Arctia caja*, (*b*) *Noctua orbona*) showing the cryptic colour pattern elements on the forewings (top arrows) and the signalling colour pattern elements on the hindwings (bottom arrows).

The relative cost-benefit trade-offs for the expression of asymmetrical cryptic colouration are therefore likely to be dependent on the ecology and species in question. Advances in digital technology and computer software will provide more powerful ways of assaying and quantifying animal symmetry by means of spectral power analysis. By using these new techniques, large meta-analyses can be constructed where large sample sizes of photographs from museum specimens are analysed simultaneously to provide a more universal picture of the principles which apply to multiple animal taxa. Other than the study by Forsman & Merilaita, there are no other studies

assaying the link between symmetry and camouflage in real species. By far the more common methodological technique when investigating such hypotheses is by designing artificial prey. Such studies have demonstrated that well established work from experimental psychology can indeed be put into a biological context. However, photographic analyses will provide a very powerful tool for determining whether these phenomena actually exist in real animals. There has been no quantification of the positioning of surface markings through the use of image analysis, which also means that an analysis of the symmetry of body patterns with respect to the axis of symmetry is yet to be investigated.

An insight into genetic and developmental constraints restraining the evolution of surface asymmetry could be better achieved by investigating the various asymmetrical body patterns of many domesticated animals, a phenomenon first identified by Wallace (1989). In domesticated animals, a breakdown of developmental canalisation through strong, artificial, directional selection indicates that genetic variation for asymmetrical colouration does exist in nature and that natural selection does indeed suppress the expression of these asymmetrical traits in the wild. A quantified comparison, perhaps using the previously described image analysis methods, could provide the scaffolding for work that would help determine the constraints opposing the evolution of asymmetrical colouration.

1.13 Concluding observations

By integrating information obtained using a diverse array of methodologies, the functional and evolutionary explanations for symmetrical animal colouration are becoming better understood. However, only after comparisons with extant species will it be possible to discover if the model patterns used in artificial target investigations have relevance in the natural world. In the context of symmetry, one of the pending questions is whether animals have evolved to optimise their surface colouration in order to reduce the intrinsic cost of being symmetrical. One of the more obvious ways this can be achieved is by placing high contrast markings away from the axis of symmetry. Perhaps a critical zone exists, similar to the one identified by the human experiments of Jenkins (1982), where zones close to the symmetry axis have a disproportional effect on symmetry detection. Whether the same principal applies to the camouflage of naturalistic targets has never been tested before. Looking into this further would improve the understanding of how animals have evolved adaptations that compensate for the developmental and genetic constraints that have arisen as part of a legacy of their evolutionary history. By using the range of approaches discussed, these hypotheses can be tested with relative ease, thus advancing research in the field of “Adaptive Colouration in Animals”, almost 80 years after Hugh Cott’s landmark book was first published.

2. The significance of the symmetry midline in animal camouflage

2.1 Introduction

Camouflage is one of the most ubiquitous visual defence strategies in the animal kingdom (Merilaita *et al.*, 2017; Nokelainen & Stevens, 2016; Stevens & Merilaita, 2009a; Stevens & Merilaita, 2011). Predation pressures have shaped the evolution of a diversity of cryptic colours and patterns (Ruxton *et al.,* 2018; Ruxton *et al.,* 2004; Stevens, 2016). However, one seemingly paradoxical feature of the patterns of most animals is the presence of bilaterally symmetrical colouration (Finnerty *et al.,* 2004; Palmer 1996). Experiments with avian predators in the field and in the laboratory have shown that symmetrically patterned camouflaged targets have lower survival rates than asymmetrical ones (Cuthill *et al.,* 2006a; Merilaita & Lind, 2006), which is consistent with earlier findings on the salience of symmetrical stimuli to humans (Barlow & Reeves, 1979; Bruce & Morgan, 1975; Gurnsey *et al.,* 1998; Rainville & Kingdom, 1999). This is because most substrates where an animal may choose to rest are asymmetrical, particularly at the spatial scale of the animal, making symmetry a salient Gestalt cue to a predator (Behrens, 1980; Behrens, 2002; Cuthill & Troscianko, 2009; Metzger, 1936; Osorio & Cuthill, 2015; Troscianko *et al.,* 2009).

Evo-devo research of the insect order Lepidoptera has suggested that there are significant genetic and developmental factors that have constrained the evolution of asymmetrical body patterns in nature (Brakefield *et al.,* 1996; Carroll *et al.,* 1994; Dilão & Sainhas, 2004; Monteiro *et al.,* 1997; Nijhout, 1991). Noteworthy exceptions in other taxa demonstrate that these constraints are not absolute (e.g. Barabás & Hancock, 1999), but decoupling surface patterning from the symmetry of the underlying morphology may require many mutations. Assuming that for most animals the benefits of reduced detectability do not outweigh these genetic and developmental costs, one might ask whether animals have evolved ways of optimising their surface patterning so as to mitigate somewhat the cost of being bilaterally symmetrical. Cuthill *et al.* (2006b) tested whether the efficacy of disruptive colouration is compromised by bilateral symmetry when using wild avian predators. They found that symmetry impeded the camouflage of background matching and disruptive strategies to similar degrees, suggesting that disruptive camouflage does not reduce the inherent cost of having symmetrical patterns. However, this field study and the laboratory work by Merilaita & Lind (2006) both indicate that the positioning of body pattern elements relative to the axis of symmetry may be of significance.

Several studies in the field of experimental psychology have demonstrated that human visual systems give a disproportionate amount of attention to features within a narrow strip of the symmetry axis (Barlow & Reeves, 1979; Bruce & Morgan, 1975; Jenkins, 1982; Julesz, 1971; Swaddle,

1999). This is most probably due to increased receiver attention at the point of fixation and so symmetry is more likely to be detected if a saccade happens to alight on the axis of symmetry (Barlow & Reeves, 1979; Locher & Nodine, 1989; Swaddle, 1999). To date, there have been no studies to test the biological relevance of this phenomenon in non-human animals, when natural-looking stimuli are used. Cuthill *et al.* (2006b) produced midline disruptive and background matching targets as a control treatment for their experiments, but the distance dependent effects of pattern elements from the axis of symmetry has never been explicitly tested in camouflage research. For a cryptic animal whose left and right elements are visible simultaneously to the receiver, one would expect camouflage adaptations, such as high contrast disruptive markings, to be placed away from the axis of symmetry. This would act as an evolutionary compromise, a strategy which enhances crypsis whilst still being bilaterally symmetrical.

The order Lepidoptera consists of approximately 180,000 described species among which there is an impressive diversity of wing colours and patterns, making them ideal model organisms for camouflage research. All species currently described display bilaterally symmetrical colouration with a clearly defined axis of symmetry, presumably because of the genetic and developmental constraints mentioned previously. The wing patterns of many species have also been shown to play a significant role in mate choice (Jiggins *et al.,* 2001; Kemp, 2007; Kemp & Rutowski, 2007; Kemp & Rutowski, 2011; Sweeney *et al.,* 2003), where in other taxa, increased levels of symmetry have been claimed to be directly linked to reproductive success (Møller, 1992, 1993; Rhodes, 2006). However, for nocturnal, cryptically coloured moths, pheromone signalling is likely to have a more significant role than colouration in intraspecific communication (Johansson & Jones, 2007). Therefore, to avoid the influence of any natural and sexual selection trade-offs, it is these camouflaged, nocturnal species that formed the focus of my work.

I wished to assess the existence of this critical region about the axis of symmetry in biologically relevant, camouflaged, artificial moth-like prey. Targets that are symmetrical nearer the axis of symmetry should appear more salient and therefore receive higher predation than targets where symmetry is placed further away from the axis. Using human visual search tasks and wild bird predation experiments, methodologies that have both been widely used in the camouflage literature (e.g. Cuthill *et al.,* 2005; Cuthill *et al.,* 2006a, 2006b; Michalis *et al.*, 2017), I wanted to determine whether these are widespread perceptual principles which apply to both mammalian and avian visual systems. As a follow-up, I then set out to test whether salient elements on the wings of real Lepidopteran species are, on average, positioned away from this critical region about the symmetry axis. This was achieved by using images from dead museum specimens and field guides, coupled with image analysis and models of salience.

2.2 Methods

2.2.1 Experiment 1: human visual search

2.2.1.1 Creation of the stimuli

Artificial moth-shaped outlines were created based on the North American moth genus *Melanolophia* (family: Geometridae), a group with a typical moth outline when in their resting position. Nine images of *Melanolophia* specimens at rest were obtained using Google Images, from which the outline was obtained by using the "lasso" tool on Paint.net.4.0.16. A perfectly symmetrical outline was created by mirroring each half of each moth specimen, giving a total of 18 shapes which acted as replicates for the different experimental treatments. The target dimensions were approximately 55 mm wide by 20 mm high with small deviations depending on the particular moth outline.

Photographs of the bark of 180 oak trees (*Quercus robor*), were taken with a calibrated Nikon D80 (Nikon Corporation, Tokyo, Japan) at Leigh Woods National Nature Reserve (North Somerset, UK, 2°38.6'W, 51°27.8'N) to provide the surface patterning of the artificial prey and the backgrounds against which they would 'rest'. That is, the prey were background-matching in the sense of being random samples from their backgrounds (Endler 1981). Each target was sampled from a different tree photo, from a randomly chosen location (selecting x and y coordinates using Matlab's random number generator for a uniform distribution). This was in order to avoid the possibility of search image formation by the human participants and, in Experiment 2, wild avian predators (e.g. Pietrewicz & Kamil, 1979). In order to create the experimental treatments, each moth was divided into sixths (three each side of the midline), a method modelled on the "six-slice" experiments of Barlow & Reeves (1979). Divisions were made by both equal area and equal width because moth shapes are not equal in area along their width, and both area and distance from the axis of symmetry could affect detectability. Five treatment groups (denoted as **S**, **I**, **M**, **O** and **A** accordingly; figure 2.1), for both division variants (equal area, **Ar**, and equal width, **W**), were created by manipulating the mirror symmetry of the bark between the complementary sixths on each half of the target. These were as follows:

Treatment S: Complete symmetry

Treatment I: Inner sixths symmetrical, the rest asymmetrical

Treatment M: Middle sixths symmetrical, the rest asymmetrical

Treatment O: Outer sixths symmetrical, the rest asymmetrical

Treatment A: Complete asymmetry

Figure 2.1. An example selection of artificial targets created from one of the *Melanolophia* specimens, showing the 5 treatment groups when divisions were made by equal area (top row) and equal width (bottom row). From left to right: **Treatment S**, **Treatment I**, **Treatment M**, **Treatment O**, **Treatment A.**

There were 18 replicates of each treatment*division combination, giving a total of 180 stimuli per trial. The targets were then placed, in the same vertical orientation, at random locations on their corresponding bark background, although they were never placed within close proximity to the location where the background for the target was extracted; these were saved as bmp files. When displayed in the experiment, the scenes were 768 pixels (20.32 cm) square, with the targets 276 (7.30 cm) pixels wide by 104 pixels (2.75 cm) high.

2.2.1.2 Running the trials

The experiment was run as a visual search task using a program written by N. Scott-Samuel and I. Cuthill (University of Bristol, UK), employing the Psychophysics Toolbox Version 3 (PTB-3) on MATLAB (The Mathworks Inc., Nattick, MA, USA) (Brainard 1997; Kleiner *et al.,* 2007), conducted from December 2017 to February 2018 using 30 human participants (50% female, 50% male, age range 18-30) with normal or corrected vision. These were recruited from the University of Bristol's School of Biological Sciences (Bristol, UK), with subjects briefed in line with the Declaration of Helsinki: the general nature of the task (but not the treatments), the anonymity of their data and right to withdraw, or withhold their data, at any point. The experiment had been approved by the University of Bristol Faculty of Science Research Ethics Committee. The visual search task was carried out on a linearized (gamma-corrected), 22'', 1024 x 768 pixel LaCie Electron 22Blue CRT monitor (LaCie Ltd., London), as in Michalis *et al.* (2017). Subjects were told to sit with their head level and within 1 m of the screen before the program was run. They were then asked to locate the moth in each of the images by clicking on its centre by using a computer touchpad (MacBook Pro; Apple Inc., Cupertino, CA, USA). Participants were given time to practice with five trial stimuli before the experiment officially began.

The 180 stimuli were displayed in a different random order for each participant, with a black fixation cross on a mid-grey background was shown for 0.5 s before each new image was displayed. Each session consisted of six blocks, each with 30 images, with the participant being given the option to take a break after each block was completed. In practice, they rarely did. Trials had a time limit of 30

s when, once reached, the program would automatically move on to the next image. Each session took approximately 25-30 minutes. Response time (RT), to the nearest 10 ms, and location of the mouse click, as well as time-outs, were recorded and the results were automatically saved as a plain text file (.txt) once each session was complete, along with information on the background filename, mask (moth outline) filename, division treatment, symmetry treatment, moth source coordinates (within the photo), and moth destination coordinates (i.e. where pasted on the background).

2.2.1.3 Analyses

Response times and residuals from fitted models were inspected visually and an inverse transform of the time (effectively response rate) was found to approximately normalise residuals. Inverse response times (1/RT) were analysed with a linear mixed model in R (R Core Team 2016) using the function lmer in the *lme4* package (Bates *et al.* 2015). Symmetry treatment (five levels) and division variant (two levels) were treated as fixed effects, whilst subject and the particular target and background used were fitted as random effects.

Visual inspection of the location of mouse clicks (Figure 2.2) shows that participants usually found the target and were highly accurate. As participants had been instructed to click anywhere on the target, accuracy analysis was attempted by taking miss/hit as a binary response variable. A miss was defined as any click outside 5% of a rectangle around the target's centre, to allow for near misses. However, owing to a low frequency of misses by participants (5.9%), the fitted models did not converge (the maximum likelihood could not reliably be found). Accuracy will therefore be ignored as a response variable. Rather than consider this a drawback, the fact that all the variation in participant performance is captured in response time is an advantage, with speed-accuracy trade-offs therefore not a concern.

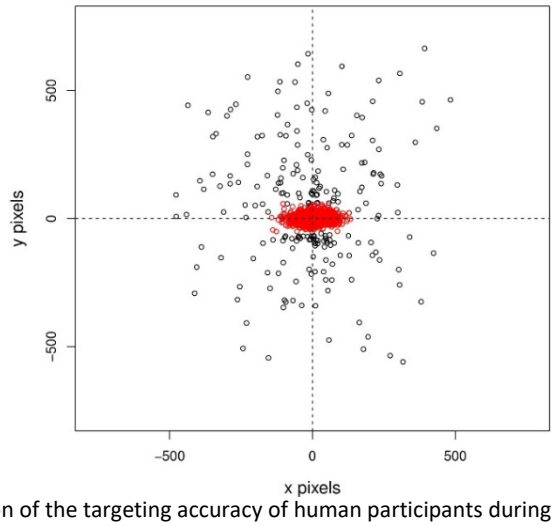


Figure 2.2. A representation of the targeting accuracy of human participants during Experiment 1, with points plotted as the distance (in pixels) from the centre of the randomly placed target. Data points in red represent hits, which were defined as any click within 5% of a rectangle around the target's centre. Due to a high proportion of hits, accuracy was ignored as a response variable.

2.2.2 Experiment 2: avian predation in the field

2.2.2.1 Procedure

The same 180 artificial prey from the human trials (see section 2.3.1.1) were calibrated by I. Cuthill (University of Bristol, UK) for passerine vision using the techniques described in Michalis et al. (2017), printed on to waterproof paper and cut out by hand to be used in the field experiment, which took place from February to March 2018. The targets were 3.5 cm wide by 2 cm tall. The experimental procedure took the same form as similar previous work investigating different aspects of animal camouflage (e.g. Cuthill *et al.* 2005). Targets were pinned on to the bark of oak trees at the mixed deciduous Leigh Woods National Nature Reserve. A dead, edible mealworm (*Tenebrio molitor* larvae frozen at -80°C then thawed) was pinned underneath each target to be used as bait, with only a small part of the mealworm being visible. A total of 1350 individual targets were put out in 15 blocks of 90, giving 8 repeats of moth shape replicates 1-9 and 7 repeats of moth shape replicates 10-18. Each block of stimuli was placed in a different area of the wood, on different dates, in order to reduce the likelihood of the replicates being exposed to the same individual predators. Fresh targets were printed and cut out for each new block.

The underside of each artificial moth was labelled with the treatment (**S**, **I**, **M**, **O** or **A**), division variant (**Ar** or **M**) and replicate number (1-18) in order to aid in mapping the location of each target in the field. Mature oak trees, usually within close proximity of footpaths, were used and each target was pinned on the side facing away from the path in order to minimise public interference. Once an individual tree was selected, individual moths were chosen at random by blindly selecting from a bag where all the targets from the particular experimental block had already been thoroughly mixed. A rough map was made to aid in localising the targets during the subsequent checks which were at 24, 48 and 72 hours.

The “survival” of each artificial moth was determined at each check by the presence or absence of the mealworm with the paper target still intact and attached to the tree. Targets were marked as “censored” if they were lost, if they survived until the end of the trial (72 h), or if there was evidence of non-avian predation. Unlike birds, which took all or most of the mealworm, arthropods such as spiders left a hollow exoskeleton, whilst molluscs such as snails left slime trails. These targets were coded as censored in the survival analysis.

2.2.2.2 Analyses

A mixed-effects Cox regression was used to perform a survival analysis on the data (as in Michalis *et al.* 2017) using the coxme function from the *coxme* R package (Therneau, 2018; R Core Team 2016).

Symmetry treatment and division variant were treated as fixed effects while experimental block was treated as a random effect. The effect of treatment was tested using an analysis of deviance which compared the unexplained variation of a model with and without the factor in question. This was tested against a χ^2 distribution for statistical significance.

2.2.3 Natural pattern analysis

2.3.3.1 Photographing, image editing, and categorisation of specimens

Dead British moth specimens were acquired from collections at the Bristol City Museum and Art Gallery (Bristol, UK, 51°45.7'N, 2°20.6'W) in May 2018. Whole drawers of the specimens were photographed using a Nikon D80 with a Nikkor 35 mm lens under controlled lighting conditions after calibration with an X-Rite ColorChecker chart (X-Rite, Tacoma Falls, USA). Scans were also taken from "British Butterflies & Moths (Collins Complete Guides)" (Sterry *et al.,* 2016), a photographic field guide which shows species in their natural resting positions. Museum and field guide specimens were then identified to family, genus, and species level (**Table 1**). Unlike the photographs taken personally, the colours in the field guide cannot be taken as accurate (even for human vision), but the rationale was that the within-wing distribution of salient features would be correlated between uncalibrated photograph and avian-perceived colour. Put simply, a species with blobs in the outer section of the wing in a photo would have an avian-perceptible blob in the outer section of the wing in real life, even if the magnitudes of the salience measures are arbitrary and could not be compared across species.

Moth images were modified using the image editing software GIMP 2. The entire body of each specimen was cropped and saved as a PNG image file. The left and right forewings were then individually cropped and, in the case of the museum specimens, rotated so that they were in the typical resting position of the species (this was judged by comparison with multiple photographs of the animal in Google Images). These were also saved as PNG image files.

Moth species of interest were ones with discrete, high contrast markings relative to the average colour and pattern of the wing. These criteria could include high contrast disruptive markings, but with the exception of horizontal markings that transected the entire wing. Only moths with such discrete markings would have had the evolutionary freedom to position pattern elements further or nearer from the axis of symmetry. In order to assign species for the analysis whilst also avoiding experimenter bias, the right forewings of the 152 total species photographed/scanned from the museum specimens and field guide were printed onto paper and cut out. The cut-outs were shuffled and shown to 20 naïve human participants, individually, under controlled conditions in November

2018. Participants were asked to place each forewing in one of two categories: one for wings with discrete, high contrast markings (Category A) and another for wings with either homogenous patterns or horizontal markings that crossed the entire wing (Category B). Participants were informed that if some of the criteria for Category A were met on any area of the wing, the wing should be placed in Category A. If no criteria for Category A were met, the wing should be placed in Category B. There was no time limit for this task to be completed but participants took approximately 15-20 minutes. Of the 152 species used during the categorisation task, 95% of the test participants put 36 of those species into category A. It was the left and right forewings of these 36 species that were then selected to enter the image analysis (figure 2.3; figure 2.4). Replicates were possible for all the moths from the museum collections. For moths obtained from the field guide, multiple images were only available for polymorphically coloured species. A phylogenetically diverse spread of moths was selected for the analysis with the sample size consisting of seven families and 31 genera (**Table 1**). Of the 36 moths chosen, nine were from the museum (figure 2.3) whilst the remaining 27 were taken from the field guide (figure 2.4).

Table 2.1 Taxonomic information on the 36 species chosen to enter the salience analysis

FAMILY	GENUS	SPECIES	COMMON NAME
DREPANIDAE	*Thyatira*	*Batis*	Peach blossom
DREPANIDAE	*Watsonalla*	*Binaria*	Oak hook-tip
EREBIDAE	*Orgyia*	*antiqua*	Vapourer
EREBIDAE	*Spilarctia*	*Luteum*	Buff ermine
GEOMETRIDAE	*Colostygia*	*pectinataria*	Green carpet
GEOMETRIDAE	*Lomographa*	*bimaculata*	White pinion spotted
GEOMETRIDAE	*Lomographa*	*temerata*	Clouded silver
GEOMETRIDAE	*Macaria*	*notata*	Peacock moth
GEOMETRIDAE	*Macaria*	*wauaria*	V-moth
GEOMETRIDAE	*Nycterosea*	*obstipata*	Gem
GEOMETRIDAE	*Opisthograptis*	*luteolata*	Brimstone moth
LASIOCAMPIDAE	*Eriogaster*	*lanestris*	Small eggar
LASIOCAMPIDAE	*Lasiocampa*	*trifolli*	Grass eggar
NOCTUIDAE	*Acontia*	*lucida*	Pale shoulder
NOCTUIDAE	*Amphipoea*	*crinanensis*	Crinan ear
NOCTUIDAE	*Amphipoea*	*lucens*	Large ear
NOCTUIDAE	*Amphipoea*	*oculea*	Ear moth
NOCTUIDAE	*Anarta*	*myrtilli*	Beautiful yellow underwing

NOCTUIDAE	*Apamea*	*ophiogramma*	Double lobed
NOCTUIDAE	*Autographa*	*bractea*	Gold spangle
NOCTUIDAE	*Autographa*	*pulchrina*	Beautiful golden Y
NOCTUIDAE	*Coranarta*	*cardigera*	Small dark yellow underwing
NOCTUIDAE	*Cosmia*	*diffinis*	White-spotted pinion
NOCTUIDAE	*Dicycla*	*oo*	Heart
NOCTUIDAE	*Dryobota*	*labecula*	Oak rustic
NOCTUIDAE	*Gortyna*	*borelii*	Fisher's estuarine moth
NOCTUIDAE	*Hadena*	*albimacula*	White spot
NOCTUIDAE	*Lacanobia*	*oleracea*	Bright-line brown eye
NOCTUIDAE	*Lenisa*	*geminipuncta*	Twin-spotted wainscot
NOCTUIDAE	*Melanchra*	*persicariae*	Dot moth
NOCTUIDAE	*Mesapamea*	*didyma*	Lesser common rustic
NOCTUIDAE	*Mesoligia*	*furuncula*	Cloaked minor
NOCTUIDAE	*Rivula*	*sericealis*	Straw dot
NOCTUIDAE	*Tyta*	*luctuosa*	Four-spotted
NOLIDAE	*Nycteola*	*revayana*	Oak nycteoline
NOTODONTIDAE	*Phalera*	*bucephala*	Buff tip

Figure 2.3 Photographic images of the moth species (9) which were taken at Bristol City Museum and Art Gallery's collections using a Nikon D80 (Nikkor 35 mm lens). After the human categorisation task, only these species were selected to enter the image analysis. Left and right forewings were cropped and rotated using GIMP2 editing software so that they were in the typical resting position of the species. From left to right: Beautiful golden Y (*Autographa pulchrina*), brimstone moth (*Opisthograptis luteolata*), buff tip (*Phalera bucephala*), dot moth (*Melanchra persicariae*), gold spangle (*Autographa bractea*), heart (*Dicycla* oo), peach blossom (*Thyatira batis*), small dark yellow underwing (*Coranarta cardigera*), white-spotted pinion (*Cosmia diffinis*). Moths are not shown to scale.

Figure 2.4 Photographic scans of the moth species (27) which were taken from the photographic field guide by Sterry *et al.* (2016), where moths were displayed in their natural resting positions (meaning that once the left and right forewings were cropped, no further adjustments were made). After the human categorizations task, only these moths were selected to enter the image analysis. From left to right: Beautiful yellow underwing (*Autographa pulchrina*), bright-line brown eye (*Lacanobia oleracea*), buff ermine (*Spilarctia luteum*), cloaked minor (*Mesoligia furunculi*), clouded silver (*Lomographa temerata*), crinan ear (*Amphipoea crinanensis*), double lobed (*Apamea ophiogramma*), ear moth (*Amphipoea oculea*), fisher's estuarine moth (*Gortyna borelii*), four-spotted (*Tyta luctuosa*), gem (*Nycterosea obstipata*), grass eggar (*Lasiocampa trifolli*), green carpet (*Colostygia pectinaria*), large ear (*Amphipoea lucens*), lesser common rustic (*Mesapamea didyma*), oak hook-tip (*Watsonalla binaria*), oak nycteoline (*Nycteola revayana*), oak rustic (*Dryobota labecula*), pale shoulder (*Acontia lucida*), peacock moth (*Macaria notata*), small eggar (*Eriogaster lanestris*), straw dot (*Rivula sericealis*), twin-spotted wainscot (*Lenisa geminipuncta*), v-moth (*Macaria wauaria*), vapourer (*Orgyia antiqua*), white pinion spotted (*Lomographa bimaculate*), white spot (*Hadena albimacula*). Moths are not shown to scale.

2.3.3.2 Image analysis

A computer program for analysing the edited right and left forewings was written by I. Cuthill using MATLAB (The Mathworks Inc., Nattick, MA, USA). The PNG files of the left and right moth forewings were read into the program, from which species and side (left or right) information was extracted. Image size was determined, and the alpha transparency channel of the PNG file ensured that only the area occupied by the wing was selected. In order to reduce the saliency of wing edges, wings were placed on a background equivalent to the mean colour (RGB) of the moth. From this, saliency maps were generated by applying an avian-vision adaptation of a model created by Rosenholtz *et al.* (2005, 2007). This is a model, used previously for investigating other aspects of animal camouflage (Xiao & Cuthill, 2016), which analyses variation in three features based on low level visual perception: luminance, colour and orientation contrast (figure 2.5). The background was replaced

with the mean colour of the wing, and the edges (3 pixels) were removed from each of the saliency maps to further reduce any effect of contrast at the edge of each wing.

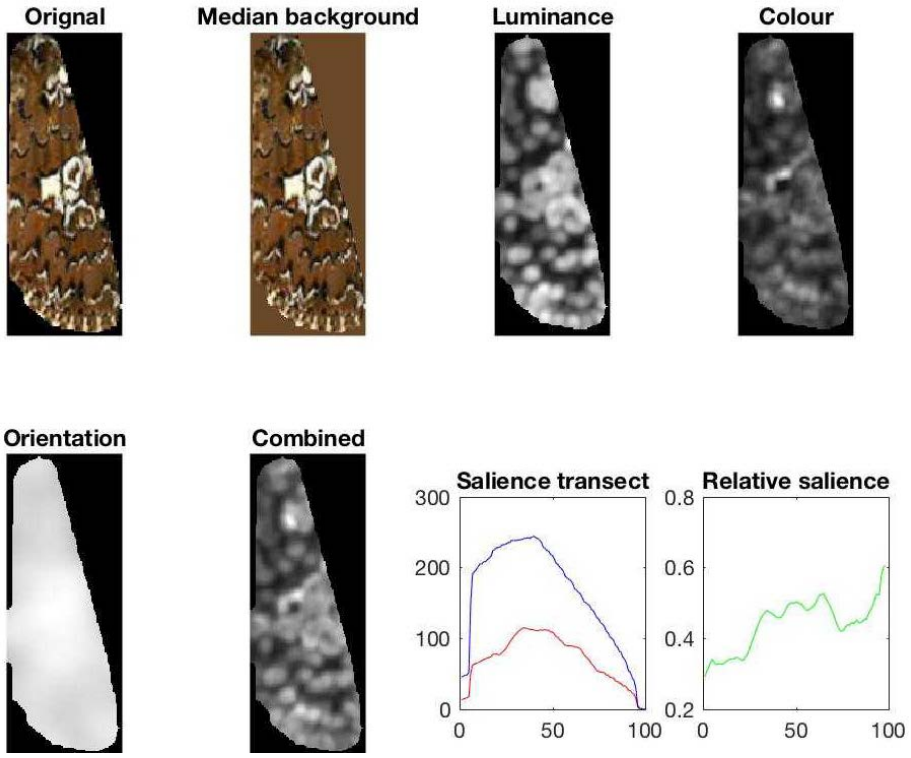


Figure 2.5 An example of the output from the wing salience computer program for the right wing of the white spot (*Hadena albimacula*), this program being an adaptation of Rosenholtz *et al.*'s (2005, 2007) code for measuring visual clutter. From left to right: the original image file, the file with background set to the median colour of the wing, a salience map based on luminance contrast, a salience map based on colour contrast, a salience map based on orientation contrast, a combined salience map, a transect along the combined salience map, showing the sum of salience values (red line) and available wing area (blue line) in each vertical column when moving from left to right along the wing, a transect of the salience score relative to the available wing area when moving from left to right along the wing.

Differences in the three salience measures (luminance, colour, and orientation) were shown to correlate well across images (figure 2.6). Luminance, colour, and orientation saliency maps were used to create a combined salience map by summing values of each column of pixels from left to right (for a right forewing) or right to left (for a left forewing). Horizontal transects going from proximal (near the midline) to distal (away from the midline) were also created and smoothed. Together, these provided a measure of the absolute salience of pattern elements across the width of the wing (figure 2.5). Because wings are not equal in area across their width, a calculation was made of relative salience, which is the proportion of the number of data points used to calculate the absolute salience at each transect point, controlling for the width of the wing at that point (figure

2.5). Species, side, absolute salience, and relative salience information was saved to a plain text file to be used in the statistical analysis.

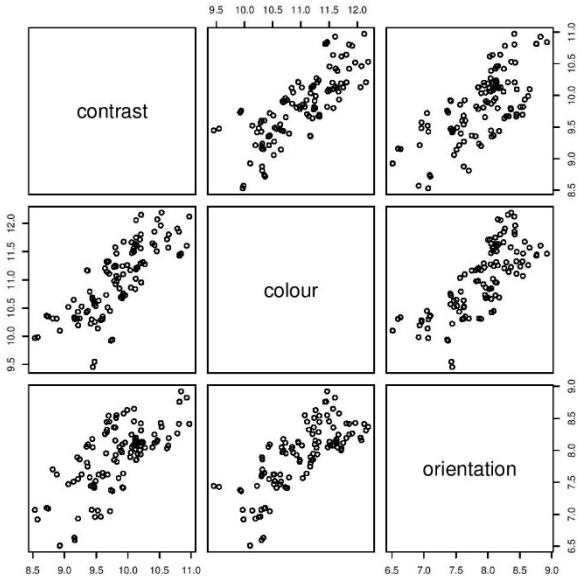


Figure 2.6 A representation of how the three salience measures (luminance contrast, colour contrast, orientation contrast) correlated across images in the natural pattern analysis

2.3.3.3 Statistical analysis

The transect order for left forewings was reversed so that the salience of both left and right sides could be analysed by moving from the proximal (midline) to the distal end of the wing. Thirds of equal width were created and the summed salience within each of these thirds was calculated. The wing area within each of these thirds was calculated and this was used to determine the relative salience (absolute salience / wing area) of each third. Within-wing differences between the middle and inner third and outer and inner third were analysed with linear mixed models in R (R Core Team 2016), using the lmer function from the *lmerTest* package (Kuznetsova *et al.,* 2017). In these models, side and species were both treated as random effects, the null hypothesis tested being that the mean difference between wing thirds is zero. The analysis was run for both absolute and relative mean salience differences. The analysis was also run (using the same random effects) according to where the image was obtained (i.e. field guide or museum).

2.3 Results

2.3.1 Experiment 1

For the inverse response times, the symmetry treatment x division variant interaction was significant (χ^2 = 13.69, d.f. = 4.0, p = 0.0084). The data were therefore split by stimuli that had been divided either by equal area (**Ar**) or equal width (**W**), with linear mixed model analyses performed separately for each. A limited set of custom contrasts was made using the glht function in the *multcomp*

package (Hothorn *et al.,* 2008) by applying a Tukey-style procedure where Treatments **S** (complete symmetry) and **A** (complete asymmetry) were contrasted with all other treatment groups.

When the targets were divided into sixths by equal area, the effect of treatment was significant (χ^2 = 256.82, d.f. = 4, p < 0.0001; figure 2.7a). There was no significant difference when Treatment **S** was contrasted with Treatment **I** (z = -0.361, p = 0.9880). However, significant differences were found when Treatment **S** was contrasted with Treatments **M** (z = 12.797, p < 0.0001), **O** (z = 3.404, p = 0.0035), and **A** (z = 12.813, p < 0.0001), where targets with Treatment **S** received a lower average response time. When Treatment **A** was contrasted with Treatment **I**, targets in Treatment **A** had a significantly greater response time (z = -4.345, p < 0.001). No significant differences were found when Treatment **A** was contrasted with Treatments **M** (z = -1.343, p = 0.5259) and **O** (z = -0.587, p = 0.9422).

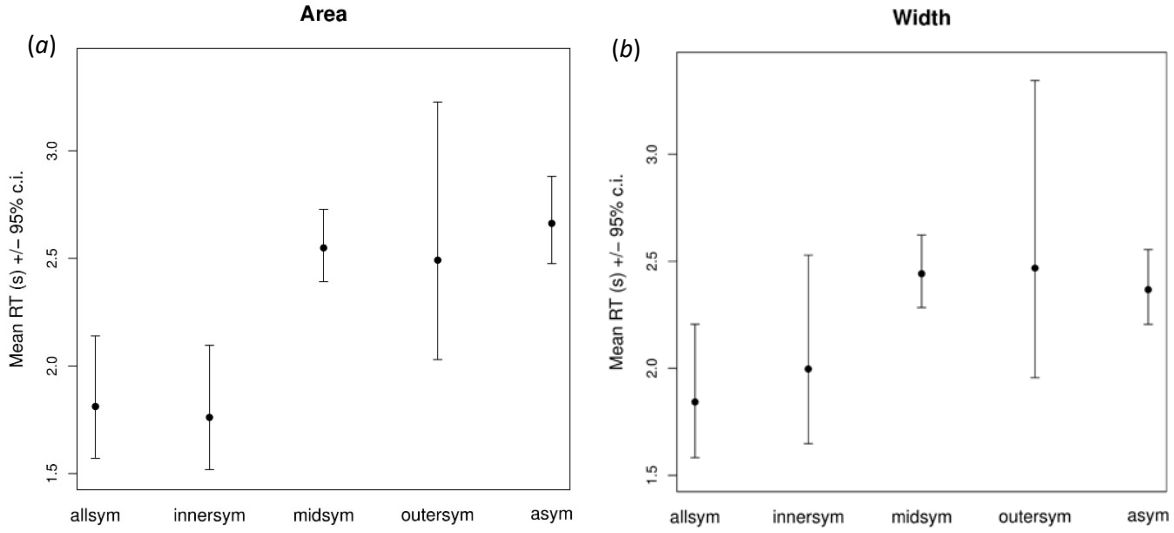


Figure 2.7 Experiment 1 results: The mean response times with respect to all treatment groups when the targets were divided by equal area *(a)* and width *(b)*. Error bars represent 95% confidence intervals. Created using the *lattice* R package (Sarkar 2008).

When the targets were divided into sixths by equal width, the effect of treatment was also significant (χ^2 = 124.65, d.f. = 4.0, p < 0.0001; figure 2.7b). As was found when targets were divided by equal area, there was also no significant difference when Treatment **S** was contrasted with Treatment **I** (z = 0.816, p = 0.8494). Again, targets with Treatment **S** received a significantly lower response time when contrasted with Treatments **M** (z = 9.705, p < 0.0001), **O** (z = 2.672, p = 0.0330), and **A** (z = 8.007, p < 0.0001). No significant differences were found when Treatment **A** was contrasted with Treatments **I** (z = -1.526, p = 0.4072), **M** (z = 0.936, p = 0.7851) and **O** (z = 0.337, p = 0.9902).

2.3.2 Experiment 2

Avian predators took 82% of all prey during the experiment. The remainder were censored, with 9% of the targets surviving and the rest being either lost (7%) or predated by spiders (1%) or slugs (1%).

The analysis of deviance from the mixed-effects Cox regression showed that the symmetry treatment x division variant interaction was not significant ($\chi^2 = 4.8441$, d.f. = 4.0, p = 0.3037) and the effect of division variant was also not significant ($\chi^2 = 0.9559$, d.f. = 1.0, p = 0.3282). The effect of symmetry treatment, however, was significant ($\chi^2 = 25.626$, d.f. = 4.0, $p < 0.0001$; figure 2.8), which led to subsequent pair-wise comparisons.

A limited set of custom contrasts was then calculated, as with the lab experiment, whereby Treatments **S** (complete symmetry) and **A** (complete asymmetry) were contrasted with all other treatment groups. When Treatment **S** was contrasted with Treatment **I**, there was no significant difference ($z = 0.438$, p = 0.9889). However, the predation rate was significantly higher for Treatment **S**, when contrasted with Treatments **M** ($z = 2.994$, p = 0.0168), and **A** ($z = 4.259$, $p < 0.0001$) (thereby confirming results by Cuthill *et al.,* 2006a). The contrast between Treatments **S** and **O** was not significant ($z = 2.576$, p = 0.0567).

Treatment **A** received a significantly lower average predation rate when contrasted with Treatment **I** ($z = -3.831$, $p < 0.0001$). No significant differences were observed when Treatment **A** was contrasted with both Treatments **M** ($z = -1.273$, p = 0.6451) and **O** ($z = -1.712$, p = 0.3610).

No birds were actually seen taking the prey, but candidate species that were frequently observed in the area were blue tits (*Cyanistes caeruleus*), great tits (*Parus major*), European robins (*Erithacus rubecula*), Eurasian nuthatches (*Sitta europaea*), Eurasian treecreepers (*Certhia familiaris*), chaffinches (*Fringilla coelebs*), and greater spotted woodpeckers (*Dendrocopos major*). Systematic observation surveys of predator species were unfeasible given the quantity of data required. When the censored data that had not survived were analysed separately, there was no significant effect of treatment ($\chi^2 = 6.843$, d.f. = 4, p = 0.1444) which is what one would expect if the complete disappearance of some of the targets was due to random factors such as wind, and if spiders and slugs had located the prey using non-visual cues or had approached the target unilaterally.

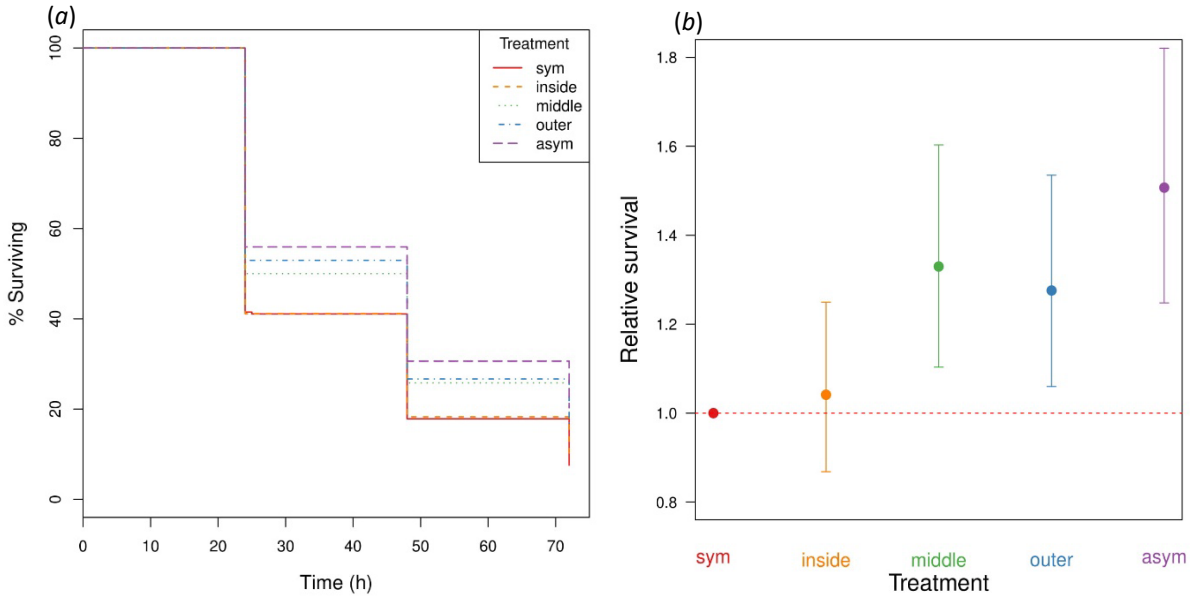


Figure 2.8 Experiment 2 results. *(a)* Survival plot with respect to treatment. A Cox regression showed that survival is best when symmetry is placed further from the midline. *(b)* Odds of survival relative to complete symmetry, the treatment which received lowest survival rate. Values and 95% confidence intervals are from the fitted survival model. Created using the *survival* and *RColorBrewer* R packages (Terry *et al.* 2000; Therneau 2000; Neuwirth 2014).

2.3.3 Natural pattern analysis

There was no significant difference between the salience of images taken from the museum collections or the field guide (t = 0.971, d.f. = 25.7, p = 0.3400), meaning that the data from all 36 moths could be used in the same analysis. The linear mixed model showed that the absolute salience of the middle third (median = 1831.8, IQR = 1238.7) was significantly greater than the absolute salience of the inner third (median = 1233.4, IQR = 821.2) (t = 9.208, d.f. = 27.2, p < 0.001). However, the inner third was significantly more salient than that of the outer third (median = 727.7, IQR = 424.8) (t = 9.687, d.f. = 35.3, p < 0.001). By necessity, the difference in salience between the middle and outer thirds was also significant (t = 11.45, d.f. = 24.7, p < 0.001) (figure 2.9; figure 2.10).

When the analysis was repeated using relative salience, both the middle third (median = 0.3631, IQR = 0.1478) and the outer third (median = 0.3941, IQR = 0.1173) had a significantly greater relative salience than the inner third (median = 0.3375, IQR = 0.1674) (t = 6.381, d.f. = 33.2, < 0.0001) (t = 5.665, d.f. = 35.2, p < 0.0001) (figure 2.9; figure 2.11). The relative salience of the middle was also significantly lower than that of the outer third, although this was not as significant as the previous pairwise comparisons (t = 2.95, d.f. = 35.3, p = 0.0060). For both absolute and relative salience measurements, the White spot (*Hadena albimacula*) displayed the greatest difference in salience between its inner and middle wing third (figure 2.12).

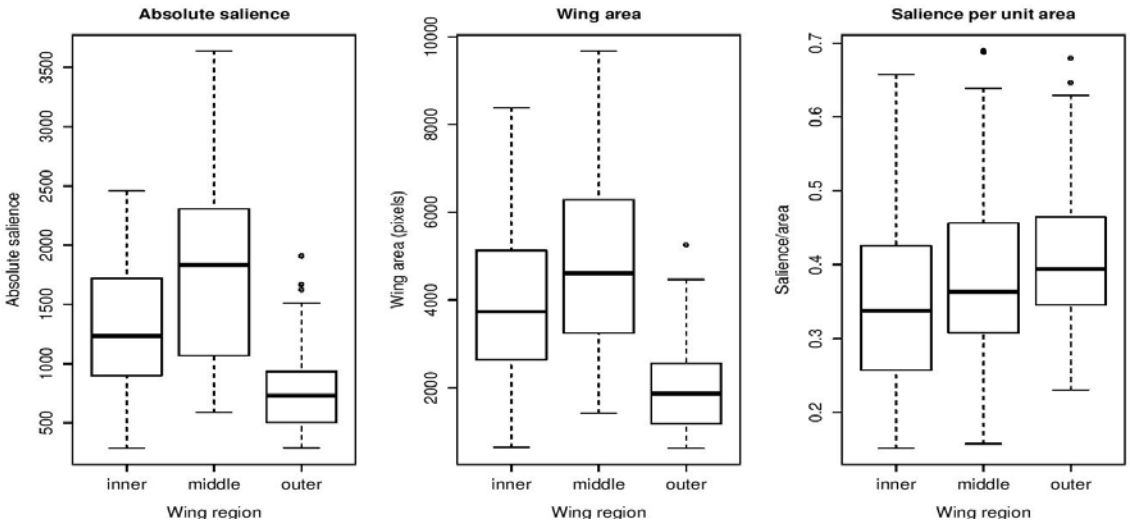


Figure 2.9 Boxplots displaying (from left to right) the absolute salience, wing area, and relative salience for the inner (closest to the midline), middle, and outer (furthest from the midline) thirds of the moth wings (natural pattern analysis) when divided by equal width. Medians (thick horizontal bars), interquartile ranges (boxes), values within 1.5 inter-quartile ranges of the box edges (whiskers) and possible outliers (circles outside the whiskers) are plotted.

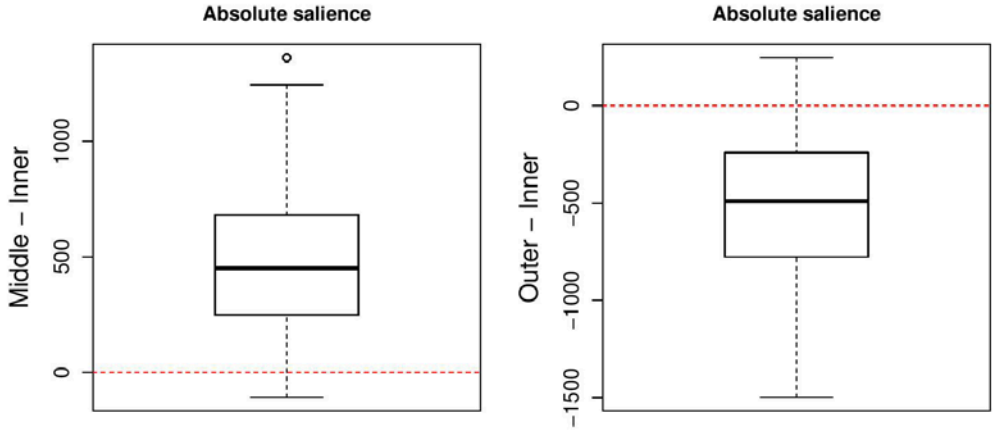


Figure 2.10 Boxplots denoting differences in absolute salience when comparing the inner and middle thirds (left) and inner and outer thirds (right) of moth wings (natural pattern analysis). Thirds were of equal width. Medians (bold line), interquartile ranges (boxes), values within 1.5 inter-quartile ranges of the box edges (whiskers) and possible outliers (circles outside the whiskers) are plotted.

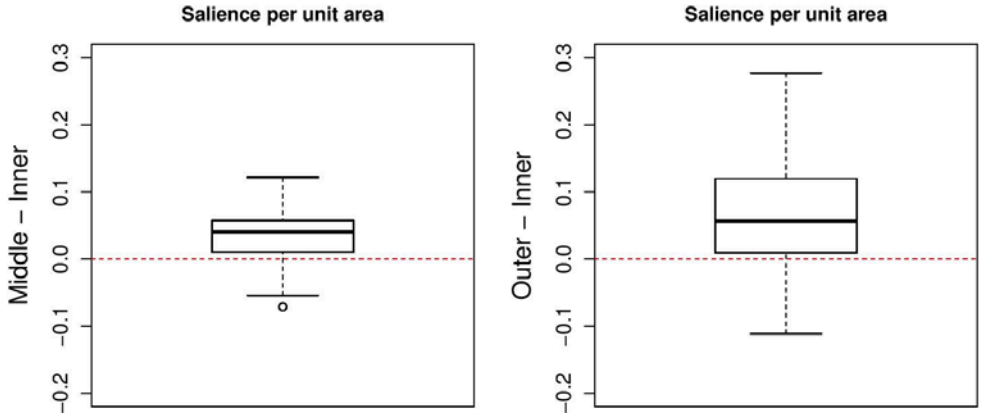


Figure 2.11 Boxplots denoting differences in relative salience when comparing the inner and middle thirds (left) and inner and outer thirds (right) of moth wings (natural pattern analysis). Thirds were of equal width. Medians (bold line), interquartile ranges (boxes), values within 1.5 inter-quartile ranges of the box edges (whiskers) and possible outliers (circles outside the whiskers) are plotted.

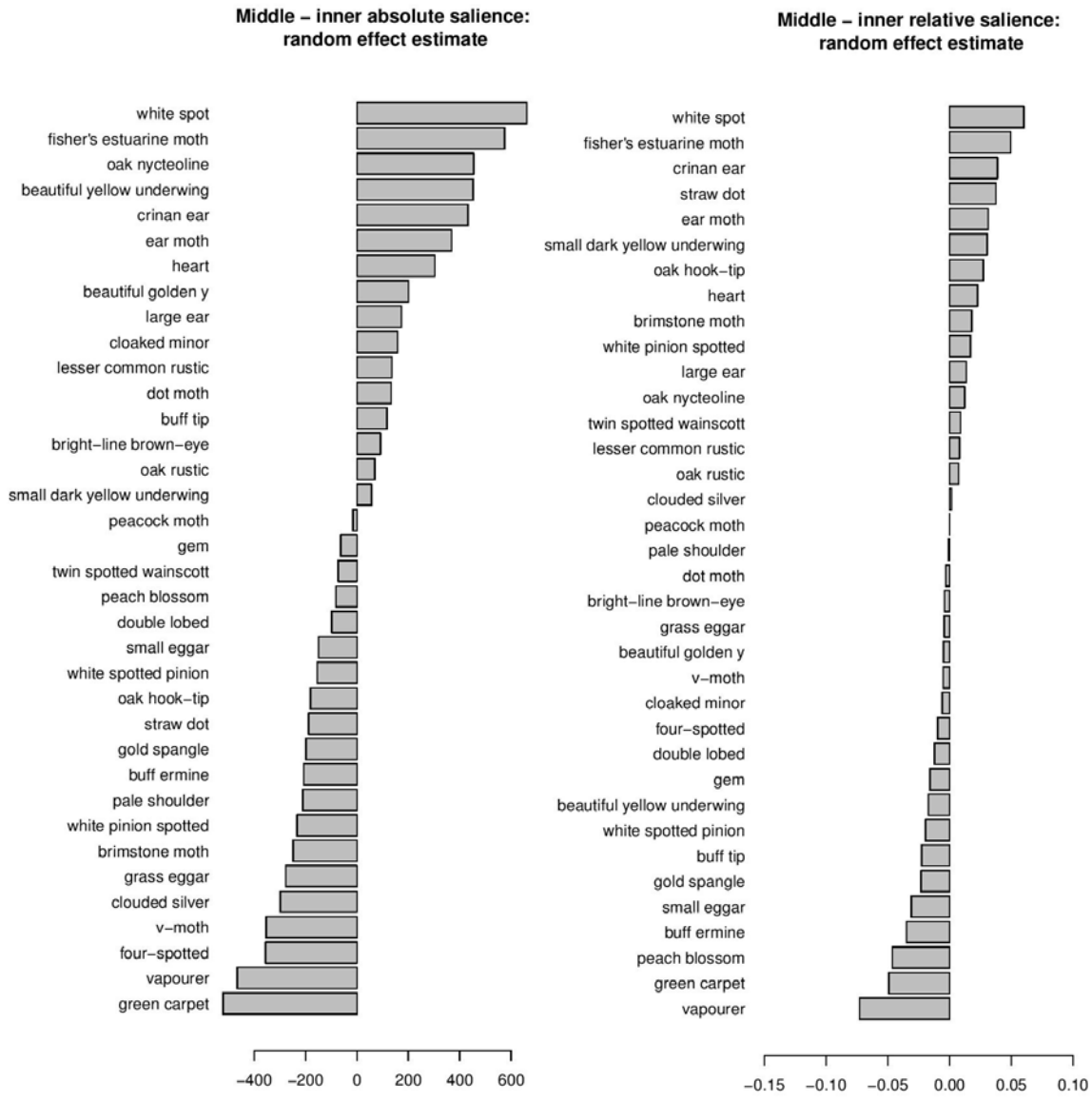


Figure 2.12 A breakdown of the direction of the median absolute (left) and relative (right) salience differences between the inner (nearest the midline) and middle thirds for each of the 36 species used in the salience analysis (natural pattern analysis).

2.4 Discussion

2.4.1 Presence of a critical zone

The results from Experiments 1 and 2 suggest the existence of a 'critical zone' surrounding the axis of symmetry of cryptically coloured animals. Prey that have symmetrical markings within this zone are more frequently detected and the output from the natural pattern analysis suggests that evolution has promoted solutions which reduce, at least to some degree, this cost in living Lepidopteran species. Jenkins (1982) used dot textures to demonstrate that the majority of symmetry information used by the human visual system is found within a strip one degree wide about the symmetry axis of the stimulus. The visual angle occupied by the inner pair of sixths in

Experiment 1 represented approximately 1.19˚ making it comparable, in terms of visual field, to the random dot stimuli used by Jenkins. By using more naturalistic stimuli, my study puts these ideas from human experimental psychology into the context of animal colours and patterns by showing that a similar phenomenon is observed in nature. The lack of significance between treatments **S** and **I** in both the human and avian experiments suggests that artificial prey which are symmetrical only near their midline are just as salient as prey which display symmetry all over their body. Now that this critical zone has been identified, its precise parameters require detailed follow-up manipulative experimentation to discover if they are indeed equivalent to the parameters found using rectangular, random dot stimuli. Furthermore, experiments on birds where the detection distance can be determined would be informative, as this factor is unknown for the field experiment presented here. The reaction times (Experiment 1) and predation rates (Experiment 2) of treatments **M** and **O** show that symmetry is relatively inconspicuous to both human and avian predators when placed outside of this critical zone (figure 2.7; figure 2.8).

2.4.2 The significance of body pattern properties

The relative cost of symmetry near the midline is likely to vary between camouflaged organisms, depending on their colouration and the nature of their respective backgrounds. Cuthill *et al.* (2006b) showed that the intrinsic cost of bilateral symmetry affected disruptive and background matching patterns to similar degrees. However, the patterns of both treatment groups in their experiments consisted of black and brown markings with identical degrees of contrast. Later findings have shown that predation rate varies when the degree of luminance contrast of disruptive markings is manipulated (Stevens *et al.,* 2006).

The marginal costs for species with homogenous background matching patterns are therefore likely to be low, whereas species with discrete, highly contrasting cryptic markings will be under stronger selection pressure to position these pattern elements away from the midline. The nature of the background or camouflage strategy will determine how many variations of positioning surface pattern elements relative to the midline are possible. It is these discrete markings that I investigated in the comparative image analysis, where I showed that in real species of Lepidoptera there is a significant tendency to place salient markings away from the midline, and that this is the case when both absolute and relative (to wing area) salience are considered. When compared with the moth wings in the natural pattern analysis, the targets used in Experiments 1 and 2 were relatively homogenous background matching patterns, created using calibrated images of oak bark. The fact that significant differences were detected using patterns where the relative marginal costs might be

low suggests that differences in Darwinian fitness between targets with discrete, contrasting markings are likely to be highly significant, and this is what the natural pattern analysis indicates.

2.4.3 Visual mechanisms

Cuthill *et al.* (2006b) did not explicitly test for differences in predation rate about the symmetry axis, but they did use midline symmetrical and non-midline symmetrical targets. No significant differences were found between these treatment groups. The authors hypothesised that midline symmetry effects did not exist owing to the large target detection distances over which wild bird predators were finding the targets. My results from Experiment 2 show that this hypothesis is in fact not true and that the positioning of symmetry relative to the midline does impact on survival (although Cuthill *et al.*'s targets were smaller by approximately 5 mm in both height and width). The results of Experiment 1 show that this is the case also when a predator is within close range of the prey item. The lack of significance in the results of Cuthill *et al.* with respect to midline might have been due to the positioning of the mealworm bait, which in my experiment was placed underneath the target rather than on top. Having the mealworm running along the top of the target midline would have highlighted the position of the axis of symmetry in all treatments and would therefore have possibly made any positioning effects of patterning undetectable during their analysis.

A symmetrical midline makes a target more salient for predators to attack because symmetrical elements are nearer each other and are therefore more easily compared by the receiver's visual system. Midline symmetry should also increase the accuracy of attack upon fixation when a predator aims to attack the centre of the prey item. The artificial targets used in Experiments 1 and 2 were similar in size to those used by Cuthill *et al.* (2006b), but Jenkins (1982) suggests that, at least for random dot studies, the range of the 'critical zone' would not change for larger prey.

As pointed out in the laboratory avian experiments by Merilaita & Lind (2006), mirror symmetry about the midline may also create abnormal patterns that are rarely present in the background, and this may provide a mechanism by which midline symmetrical prey are more efficiently detected. Although this may have influenced the results from Experiments 1 and 2, the complex oak bark texture of the targets is likely to have suppressed its effects in comparison with the highly salient, unnatural-looking, chevron patterns generated by Merilaita & Lind in their captive great tit experiment. The idea of rare pattern generation and the alternative mechanism, that symmetry within the critical zone is more salient owing to pattern elements being closer to the point of fixation, are not mutually exclusive. However, abnormal patterns were not encountered in the random dot trials conducted by human experimental psychologists (Barlow & Reeves, 1979, Jenkins, 1982) because of the nature of the stimuli used. The relative effect of rare pattern generation on

saliency is therefore likely to vary depending on the type of pattern, but the idea that midline symmetrical targets have markings closer to the point of fixation of a receiver should apply to all bilaterally symmetrical, camouflaged animals.

2.4.4 Comparisons between Experiments 1 and 2

Having to divide the artificial targets into equal segments by both area and width was not something that Barlow & Reeves' (1979) "six-slice experiment" needed to factor in. The rectangular nature of their stimuli meant divisions by equal area and equal width were equivalent. In the human experiment, division variant was shown to have a significant effect on stimulus detection whereby Treatments **S** and **I** received more similar response times when divided by equal area. The area occupied by the inner third of treatment **I** was approximately 43% greater when divided by equal width than for equal area so this may explain these differences in response time when viewed at close range by the human participants. During Experiment 2, there was no significant difference between division variants, presumably because during Experiment 1 the human predators were within far closer visual range of the prey items, which made differences between the two variants more obvious. Nevertheless, the effect of treatment showed a very similar trend in both variants of the lab experiment to the field experiment. It would be interesting to conduct complementary experiments with captive birds in the laboratory (e.g. Merilaita & Lind, 2006) and detection trials with humans in the field (e.g. Xiao & Cuthill, 2016), using the same targets to ascertain if this would give complementary results to the findings presented here.

Fig. 2.7 shows that for Experiment 1, the 95% confidence intervals for Treatment **O** were larger than for the other treatment groups, in both width and area variants. The reasons for this increased variation could be because the outer third contained a larger proportion of the perimeter of the target (as in Barlow & Reeves, 1979). On average, 43% of the target perimeter was within the outer third but this average varied between division variant **Ar** (51%) and **W** (36%). The extent to how this might have impacted the results of Experiment 1 may have been variable owing to the random positioning of the stimulus on the screen and the particular oak bark background used (which was different for each of the 180 stimuli per trial). Therefore, all things being equal, symmetry in outline (as opposed to surface) should be more detectable for Treatment **O** than for any of the other treatments although empirical investigations with biologically relevant stimuli are lacking.

Enhanced variation of predation on Treatment **O** was not observed in the results of Experiment 2, presumably again owing to the larger distances at which targets were detected by wild birds in the woodland. This could explain the lack of significance when Treatment **S** was contrasted with

Treatment **O** in Experiment 2 but the fact that not all variables could be controlled in the field provides reasons to predict statistical power to be lower in the avian predation experiment anyway.

Another unusual finding during Experiment 1 was the lack of significance between Treatment **A** and Treatment **I** when targets were divided by equal width. Again, the proximity of the participants to the target during each trial is the only likely explanation. Predation by naïve predators under natural conditions did not replicate these results, suggesting that this is again an artefact of the visual search task's design.

2.4.5 Body orientation

Outcomes from both the human and avian experiments confirm previous findings by Cuthill *et al.* (2006a) and Merilaita & Lind (2006) that symmetrical colouration decreases the efficacy of crypsis. Both studies used predators which would normally view prey from an angle where the bilateral symmetry of the target would be obvious to the receiver. This was also the case for all three experiments presented here. All moth species chosen for the Natural Pattern Analysis occupied resting positions that made both forewings simultaneously visible (figure 2.3; figure 2.4). As in our experiments, Cuthill *et al.* (2006a, 2006b) and Merilaita & Lind (2006) also used targets that positioned the axis of symmetry in a vertical orientation, which is where symmetry is more efficiently discriminated (Corballis & Roldan, 1975). The relative cost of bilateral symmetry on crypsis is therefore likely to be dependent on orientation. This might explain the seemingly contradictory results obtained by Langridge (2006), who showed that the dynamic colouration of the cuttlefish *Sepia officinalis* was in fact more symmetrical when displaying camouflaged patterns as opposed to deimatic patterns. For a marine predator hunting from above, the orientation of the axis of symmetry would probably not be in the vertical direction, and thus the unpredictability of midline orientation might interfere with a predator's perception of symmetry (see section 1.7 for more details).

Body orientation has indeed been shown to have a significant effect on the detectability of cryptic prey (Pietrewicz & Kamil, 1977), and this has been supported with evidence from living Lepidoptera, where resting orientation was shown to be non-random and varied depending on the surface patterning of the species and the nature of the background (Webster *et al.,* 2009). Behaviourally mediated crypsis may therefore provide another solution for optimising camouflage, and there is now evidence of behavioural choice of body orientation in Lepidoptera not necessarily in the vertical plane (Kang *et al.,* 2012). These prey items are therefore likely to evolve different body patterns to species which would normally rest vertically on substrates such as tree bark. Nevertheless, we should still expect selection for high contrast markings to be positioned away from the 'critical zone'

near the axis of symmetry, as placing elements away from this area should still reduce detectability, regardless of orientation.

During the moth categorization task in the Natural Pattern Analysis, participants were told to choose wings with high contrast, vertical, disruptive markings. Given the findings by Webster *et al.* (2009), these species should in any case have an increased probability of resting in the vertical plane as this would increase the efficacy of background matching. Resting orientation is therefore an aspect which should be taken into consideration when trying to quantify the relative cost of symmetrical colouration, but it lacks empirical field investigation. The relationship between body orientation and symmetry perception has only been explicitly demonstrated using random dot stimuli and human participants. Studies with more naturalistic targets and non-human predators are required for these experimental psychological phenomena to be generalised to other taxa. The symmetry perceptual capabilities of the predators in question should also be investigated in more detail.

2.4.6 Constraints due to wing area

The image analysis of museum and field guide specimens suggests that camouflaged Lepidoptera have evolved to place highly contrasting, cryptic pattern elements away from the inner third, nearest to the midline. When comparing values of absolute salience, there was significantly higher salience in the middle third than in the inner third, but the salience from the inner third was significantly higher than from the outer third (figure 2.9; figure 2.10). This is primarily because there is less wing area in the outer third to contribute to the total salience score and this was demonstrated when the analysis was re-run using relative salience (figure 2.6; figure 2.11). Both the middle and outer thirds had greater salience relative to the wing area than the inner third. The shape of Lepidopteran wings therefore adds to the intrinsic cost of symmetrical colouration by virtue of there being more area near the midline.

Given that the highly salient markings of interest included disruptive colourations - patterns that break the body outline - it is possible that these were more likely to evolve on the outer third given that it was more likely to contain a relatively greater proportion of the wing edge. This was not specifically tested, although the perimeter of each wing (3 pixels) was ignored when calculating the salience count of each vertical column, which might have suppressed its effect. Observations of the moth species chosen for the experiment (figure 2.3, figure 2.4) show that 18 of the 36 species do not in any case have disruptive markings near the wing edge.

Even though the median wing area is greatest in the middle third, my results demonstrate that the saliency of pattern elements relative to wing area is still greater in the middle third than in the inner

third. Absolute values of saliency are ultimately more relevant in the context of the visual ecologies of predators searching for these prey items against their respective backgrounds. However, what emerges from the analysis of relative salience is that, despite the constraint of wing shape, animals have evolved to place a disproportionally high number of salient markings away from their body midline.

2.4.7 Alternative explanations for enhanced salience away from the midline

An alternative adaptive explanation for the existence of highly contrasting markings away from the midline would be for a so-called "distractive effect" (Thayer, 1909), which diverts a predator's attention away from the outline of the prey item. There is considerable disagreement in the literature on the functional significance of alleged distractive markings as camouflage (Dimitrova *et al.,* 2008; Merilaita *et al.,* 2013; Stevens *et al.,* 2008; Stevens *et al.,* 2013a; Stevens *et al.,* 2013b), but these authors have used Lepidopteran species as potential examples. Experiments undertaken under natural conditions have suggested that distractive markings do not enhance survival, but more research is required in order to establish the circumstances in which camouflage by distraction could occur, if indeed it occurs at all. Knowledge of the characteristics of the background would have been required for us to justifiably assess the relative significance of distractive markings in the species studied in the pattern analysis. This was beyond the scope of the project.

The functional significance of eyespots has usually been considered in the context of diverting attack, and this has been empirically demonstrated to be the case in experiments with sticklebacks (Kjernsmo & Merilaita, 2013; Kjernsmo *et al.,* 2016) and mantises (Prudic *et al.,* 2015) as predators. However, indicators of the presence of eyespots (concentric rings of contrasting colours) were not observed in any of the 36 species. Moths which did have eyespot characteristics on their forewings were discounted even before the human categorization task, as moths with these patterns would not have been classed as being camouflaged.

2.4.8 The significance of phylogeny

Although the species selected for the pattern analysis were not analysed with respect to phylogeny, a detailed examination of the phylogenetic relationships of each of the species studied might throw more light on the adaptive significance of placing pattern elements away from the 'critical zone' and might put into context the variation that is observed between the species in our studies (figure 2.12) (not all the species followed the trend, but figures 2.10 and 2.11 show that the majority did). Therefore, the phenomenon of placing salient markings away from the midline is quite likely to be

present in other camouflaged animals that rest on their respective backgrounds with their left and right sides simultaneously visible.

2.4.9 Conclusions and future work

My work provides insights into how camouflaged prey have evolved to suppress, at least in part, the predation cost due to symmetrical colour patterns. Experiments 1 and 2 show that the effect of symmetry near the midline has similar effects in both mammalian and avian visual systems, whilst the pattern analysis of real moths demonstrates that this phenomenon is indeed observed in a broad phylogenetic spread of living Lepidopteran species. Image analyses of this kind are rare in the camouflage literature, where most work has centred around designing experiments using artificial prey that are normally not based on any particular species found in nature. Forsman & Merilaita (2003) quantified the symmetry on the cryptic forewings and signalling hindwings of three species of moth by making morphological measurements of individual pattern elements (see section 1.12.3), a technique that is not comparable to the methods applied in this thesis. It is hoped that advances in image analysis and geometric morphometric technologies will increase the frequency of application of these techniques to the field of animal colouration, thus helping to put the results from artificial target experiments into a natural historical context.

The relative importance of genetics and development in constraining the evolution of asymmetrical colouration remains unclear. Nevertheless, one should assume that evolving high contrast pattern elements away from the midline to be developmentally simpler than evolving totally bilaterally asymmetrical patterns. Evo-devo research using models such as *Tithrone roseipennis* (Barabas & Hancock, 1999), a species of mantis that has asymmetrically coloured hindwings, could help quantify the importance of pleiotropy and other effects that might be constraining the evolution of asymmetry in most other animal species. The use of CRISPR/Cas9 somatic mutagenesis in creating loss-of-function mutants in gene families such as *Wnt* has highlighted how minor changes in gene expression can generate significant morphological changes (Martin & Reed, 2017; Mazo-Vargas *et al.,* 2017). The possibility of quantifying these effects on discrete wing traits is only beginning to receive attention. F_2 hybrid crosses and QTL mapping techniques would enable correlations to be made between the observed phenotype and allelic frequency in a similar way to how other functional genes under selection have been identified in the past (e.g. Colosimo *et al.,* 2005; Steiner *et al.,* 2007). The advantage of this would be to enable monitoring selection on asymmetrical colouration in the wild by using living animal models.

The results presented here have therefore generated a wealth of follow-up questions which require further investigation. The specific parameters of the 'critical zone' require detailed quantification

(section 2.5.1). Complementary human field and avian lab experiments should be undertaken to extend the results of Experiments 1 and 2 (section 2.5.4). The effect of body orientation with respect to symmetry perception should be studied within the context of camouflage (section 2.5.5). An enhanced knowledge of the potential significance of distractive markings would aid our interpretation of the adaptive importance of salient markings away from the midline (section 2.5.7). These are some of the follow-up questions which would contribute to deepening our knowledge of the evolutionary ecology of symmetrical colouration with respect to camouflage and would, in the process, enhance our understanding of predator-prey interactions.

References

Adamo, S. A., Ehgoetz, K., Sangster, C. & Whitehorne, I. (2006). Signalling to the enemy? – Body pattern expression and its response to external cues during hunting in the cuttlefish *Sepia officinalis* (Cephalopoda), *Biological Bulletin*, **210**, 192-200.

Allen, J. J., Mathger, L. M., Barbosa, A., Buresch, K. C., Sogin, E., Schwartz, J., Chubb, C., & Hanlon, R. T. (2010). Cuttlefish dynamic camouflage: responses to substrate choice and integration of multiple visual cues, *Proceedings of the Royal Society B Biological Sciences*, **277**, 1031-1039.

Allen, J. J., Mathger, L. M., Barbosa, A. & Hanlon, R. T. (2009). Cuttlefish use visual cues to control three-dimensional skin papillae for camouflage, *Journal of Comparative Physiology A-Neuroethology Sensory Neural and Behavioral Physiology,* **195**, 547-555.

Badyaev, A. V., Hill, G. E., Dunn, P. O., & Glen, J. C. (2001). Plumage color as a composite trait: Developmental and functional integration of sexual ornamentation, *American Naturalist*, **158**, 221-235.

Barabás, S. P. & Hancock, E. G. (1999). Asymmetrical colour and wing-folding in *Tithrone roseipennis* (Saussure 1870) a neotropical praying mantis (Mantodea Hymenopodidae), *Tropical Zoology*, **12**, 325-334.

Barbosa, A., Litman, L. & Hanlon, R. T. (2008). Changeable cuttlefish camouflage is influenced by horizontal and vertical aspects of the visual background, *Journal of Comparative Physiology A-Neuroethology Sensory Neural and Behavioral Physiology,* **194**, 405-413.

Barbosa, A., Mathger, L. M., Buresch, K. C., Kelly, J., Chubb, C., Chiao, C. C. & Hanlon, R. T. (2008). Cuttlefish camouflage: The effects of substrate contrast and size in evoking uniform, mottle or disruptive patterns, *Vision Research*, **48**, 1242-1253.

Barlow, H. B. & Reeves, B. C. (1979). Versatility and absolute efficiency of detecting mirror symmetry in random dot displays, *Vision Research*, **19**, 783-793.

Barnett, J. B., Cuthill, I. C. & Scott-Samuel, N. E. (2017). Distance-dependent pattern blending can camouflage salient aposematic signals, *Proceedings of the Royal Society B-Biological Sciences*, **284**, Article no. 20170128.

Bates, D., Maechler, M., Bolker, B. & Walker, S. (2015). Fitting linear mixed-effects models using lme4, *Journal of Statistical Software*, **67.1**, 1-48 (doi:10.18637/jss.v067.i01).

Behrens, R. R. (1980). Camouflage, Art And Gestalt. *North American Review,* **265,** 8-18.

Behrens, R. R. (2002). *False Colors: Art, Design and Modern Camouflage,* Dystart, Iowa, Bobolink Books.

Brainard, D. H. (1997). The psychophysics toolbox, *Spatial Vision*, **10**, 433-436.

Brakefield, P. M., Gates, J., Keys, D., Kesbeke, F., Wijngaarden, P. J., Monteiro, A., French, V. & Carroll, S. B. (1996). Development, plasticity and evolution of butterfly eyespot patterns, *Nature*, **384**, 236-242.

Breuker, C. J. & Brakefield, P. M. (2002). Female choice depends on size but not symmetry of dorsal eyespots in the butterfly *Bicyclus anynana*, *Proceedings of the Royal Society B-Biological Sciences*, **269**, 1233-1239.

Bruce, V. G. & Morgan, M. J. (1975). Violations of symmetry and repetition in visual-patterns, *Perception*, **4**, 239-249.

Buresch, K. C., Maethger, L. M., Allen, J. J., Bennice, C., Smith, N., Schram, J., Chiao, C. C., Chubb, C. & Hanlon, R. T. (2011). The use of background matching vs. masquerade for camouflage in cuttlefish *Sepia officinalis*, *Vision Research*, **51**, 2362-2368.

Carroll, S. B., Gates, J., Keys, D. N., Paddock, S. W., Panganiban, G. E. F., Selegue, J. E. & Williams, J. A. (1994). Pattern formation and eyespot determination in butterfly wings, *Science*, **265**, 109-114.

Cheverud, J. M. (1984). Quantitative genetic and developmental constraints on evolution by selection, *Journal of Theoretical Biology*, **110**, 155-171.

Chiao, C. C., Chubb, C., Buresch, K., Siemann, L. & Hanlon, R. T. (2009). The scaling effects of substrate texture on camouflage patterning in cuttlefish, *Vision Research*, **49**, 1647-1659.

Chiao, C. C., Kelman, E. J. & Hanlon, R. T. (2005). Disruptive body patterning of cuttlefish (*Sepia officinalis*), *Biological Bulletin*, **208**, 7-11.

Chlao, C. C., Chubb, C. & Hanlon, R. T. (2007). Interactive effects of size, contrast, intensity and configuration of background objects in evoking disruptive camouflage in cuttlefish, *Vision Research*, **47**, 2223-2235.

Colosimo, P. F., Hosemann, K. E., Balabhadra, S., Villarreal, G., Dickson, M., Grimwood, J., Schmutz, J., Myers, R. M., Schluter, D. & Kingsley, D. M. (2005). Widespread parallel evolution in sticklebacks by repeated fixation of ectodysplan alleles, *Science*, **307**, 1928-1933.

Corballis, M. C. & Roldan, C. E. (1975). Detection of symmetry as a function of angular orientation, *Journal of Experimental Psychology – Human Perception and Performance*, **1**, 221-230.

Cott, H. B. (1940). *Adaptive Coloration in Animals,* London, Methuen & Co. Ltd.

Cuthill, I. C., Stevens, M., Sheppard, J., Maddocks, T., Párraga C. A. & Troscianko, T. S. (2005). Disruptive coloration and background pattern matching, *Nature*, **434**, 72-74.

Cuthill, I. C., Hiby, E. & Lloyd, E. (2006). The predation costs of symmetrical cryptic colouration, *Proceedings of the Royal Society B Biological Sciences*, **273**, 1267-1271.

Cuthill, I. C., Stevens, M., Windsor, A. M. M. & Walker, H. J. (2006). The effects of pattern symmetry on detection of disruptive and background-matching coloration, *Behavioral Ecology*, **17**, 828-832.

Cuthill, I. C. & Szekely, A. (2009). Coincident disruptive coloration, *Philosophical Transactions of the Royal Society B-Biological Sciences*, **364**, 489-496.

Cuthill, I. C. & Troscianko, T. S. (2009). Animal camouflage: biology meets psychology, computer science and art. *International Journal of Design & Nature and Ecodynamics,* **4,** 183-202.

Cuthill, I. C., Troscianko, T. S., Kibblewhite, A., King, O. & Stevens, M. (2007). Edge enhancement in disruptive camouflage. *Perception,* **36**, 1399-1399.

Darwin, C. (1859). *On the Origin of Species by Means of Natural Selection, or the Preservation of Favoured Races in the Struggle for Life.,* London, John Murray.

Delius, J. D. & Habers, G. (1978). Symmetry – Can pigeons conceptualize it?, *Behavioral Biology*, **22**, 336-342.

Dimitrova, M. & Merilaita, S. (2010) Prey concealment: visual background complexity and prey contrast distribution, *Behavioral Ecology,* **21**, 176-181.

Dimitrova, M. & Merilaita, S. (2012). Prey pattern regularity and background complexity affect detectability of background-matching prey, *Behavioral Ecology*, **23**, 384-390.

Dimitrova, M., Stobbe, N., Schaefer, H. M. & Merilaita, S. (2009). Concealed by conspicuousness: distractive prey markings and background, *Proceedings of the Royal Society B-Biological Sciences,* **276**, 1905-1910.

Endler, J. A. (1981). An overview of the relationships between mimicry and crypsis, *Biological Journal of the Linnean Society*, **16**, 25-31.

Enqvist, M. & Arak, A. (1994). Symmetry, beauty and evolution, *Nature*, **372**, 169-172.

Espinosa, I. & Cuthill, I. C. (2014). Disruptive colouration and perceptual grouping, *PLOS ONE,* **9**, Article no. e87153.

Evans, C. S., Wendeworth, P. & Cheng, K. (2000). Detection of bilateral symmetry in complex biological images, *Perception*, **29**, 31-42.

Finnerty, J. R., Pang, K., Burton, P., Paulson, D. & Martindale, M. Q. (2004). Origins of bilateral symmetry: Hox and dpp expression in a sea anemone, *Science*, **304**, 1335-1337.

Forsman, A. & Merilaita, S. (1999). Fearful symmetry: pattern size and asymmetry affects aposematic signal efficacy, *Evolutionary Ecology*, **13**, 131-140.

Forsman, A. & Merilaita, S. (2003). Fearful symmetry? Intra-individual comparisons of asymmetry in cryptic vs. signalling colour patterns in butterflies, *Evolutionary Ecology*, **17**, 491-507.

Forsman, A. & Herrstrom, J. (2004). Asymmetry in size, shape, and color impairs the protective value of conspicuous color patterns, *Behavioral Ecology*, **15**, 141-147.

Giurfa, M., Eichmann, B. & Menzel, R. (1996). Symmetry perception in an insect, *Nature*, **382**, 458-461.

Giurfa, M., Dafni, A. & Neal, P. R. (1999). Floral symmetry and its role in plant-pollinator systems, *International Journal of Plant Sciences*, **29**, 345-373.

Guilford, T. (1990). The secrets of aposematism – Unlearned responses to specific colours and patterns, *Trends in Ecology & Evolution*, **5**, 323-323.

Gurnsey, R., Herbert, A. M. & Kenemy, J. (1998). Bilateral symmetry in noise is detected accurately only at fixation, *Vision Research*, **38**, 3795-3803.

Hanlon, R. T. (2007). Cephalopod dynamic camouflage, *Current Biology*, **17**, 400-404.

Hanlon, R. T. & Messenger, J. B. (1988). Adaptive coloration in young cuttlefish (*Sepia officinalis* L): The morphology and development of body patterns and their relation to behaviour, *Philosophical Transactions of the Royal Society of London Series B-Biological Sciences*, **320**, 437-487.

Hanlon, R. & Messenger, J. B. (2018). *Cephalopod Behaviour (Second Edition)*, Cambridge, UK, Cambridge University Press.

Hanlon, R., Vecchione, M. & Allcock, L. (2018). *Octopus, Squid & Cuttlefish: The worldwide illustrated guide to cephalopods,* Chicago, University of Chicago Press.

Hanlon, R. T., Watson, A. C. & Barbosa, A. (2010). A "Mimic Octopus" in the Atlantic: Flatfish mimicry and camouflage by *Macrotritopus defilippi*, *Biological Bulletin*, **218**, 15-24.

Hoekstra, H. E. (2006). Genetics, development and evolution of adaptive pigmentation in vertebrates, *Heredity*, **97.3**, 222-234.

Hollard, V. D. & Delius, J. D. (1982). Rotational invariance in visual pattern recognition by pigeons and humans, *Science*, **218**, 804-806.

Hopkins, T. L. & Kramer, K. J. (1992). Insect cuticle sclerotization, *Annual Review of Entomology*, **37**, 273-302.

Horridge, G. A. (1996). The honeybee (*Apis mellifera*) detects bilateral symmetry and discriminates it axis, *Journal of Insect Physiology*, **42**, 755-764.

Hothorn, T., Bretz, F. & Westfall, P. (2008). Simultaneous inference in general parametric models, *Biometrical Journal*, **50.3**, 346-363.

Jenkins, B. (1982). Redundancy in the perception of bilateral symmetry in dot textures, *Perception & Pyschophysics*, **32**, 171-177.

Jiggins, C. D., Naisbit, R. E., Coe, R. L. & Mallet, J. (2001). Reproductive isolation caused by colour pattern mimicry, *Nature*, **411**, 302-305.

Johansson, B. G. & Jones, T. M. (2007). The role of chemical communication in mate choice, *Biological Reviews*, **82**, 265-289.

Johnstone, R. A. (1994). Female preference for symmetrical males as a by-product of selection for mate recognition, *Nature*, **372**, 172-175.

Julesz, B. (1971). *Foundations of cyclopean perception*. University of Chicago Press.

Kang, C. -K., Moon, J. Y., Lee, S. I. & Jablonski, P. G. (2012). Camouflage through an active choice of a resting spot and body orientation in moths, *Journal of Evolutionary Biology,* **25**, 1695-1702.

Kelman, E. J., Tiptus, P. & Osorio, D. (2006). Juvenile plaice (*Pleuronectes platessa*) produce camouflage by flexibly combining two separate patterns, *Journal of Experimental Biology*, **209**, 3288-3292.

Kemp, D. J. (2007). Female butterflies prefer males bearing bright iridescent ornamentation, *Proceedings of the Royal Society B-Biological Sciences,* **274**, 1043-1047.

Kemp. D. J. & Rutowski, R. L. (2011). The role of coloration in mate choice and sexual interactions in butterflies, *Advances in the Study of Behavior*, **43**, 55-92.

Kjernsmo, K., Gronholm, M. & Merilaita, S. (2016). Adaptive constellations of protective marks: eyespots, eye stripes and diversion of attacks by fish, *Animal Behaviour*, **111**, 189-195.

Kjernsmo, K. & Merilaita, S. (2013). Eyespots divert attacks by fish, *Proceedings of the Royal Society B-Biological Sciences,* **280**, Article no. 20141458.

Kleiner, M. Brainard, D. & Pelli, D. (2007). What's new in Psychtoolbox-3?, *Perception*, **36(Suppl.)**, 1-16.

Kuznetsova, A., Brockhoff, P. B & Christensen, R. H. B. (2017). lmerTest package: tests in linear mixed effects models, *Journal of Statistical Software,* **82**, 1-26. doi: 10.18637/jss.v082.i13 (URL: http://doi.org/10.18637/jss.v082.i13).

Langridge, K. V. (2006). Symmetrical crypsis and asymmetrical signalling in the cuttlefish *Sepia officinalis*, *Proceedings of the Royal Society B Biological Sciences*, **273**, 959-967.

Langridge, K. V., Broom, M. & Osorio, D. (2007). Selective signalling by cuttlefish to predators, *Current Biology*, **17**, 1044-1045.

Locher, P. & Nodine, C. (1989). The perceptual value of symmetry, *Computers & Mathematics with Applications*, **17**, 475-484.

Kelman, E. J., Baddeley, R. J., Shohet, A. J. & Osorio, D. (2007). Perception of visual texture and the expression of disruptive camouflage by the cuttlefish, *Sepia officinalis*, *Proceedings of the Royal Society B-Biological Sciences*, **274**, 1369-1375.

Maethger, L. M., Chiao, C. C., Barbosa, A., Buresch, K. C., Kaye, S. & Hanlon, R. T. (2007). Disruptive coloration elicited on controlled natural substrates in cuttlefish, *Sepia officinalis*, *Journal of Experimental Biology*, **210**, 2657-2666.

Mappes, J., Kokko, H., Ojala, K. & Lindstrom, L. (2014). Seasonal changes in predator community switch the direction of selection for prey defences, *Nature Communications*, **5,** Article no. 5016.

Mappes, J., Marples, N. & Endler, J. A. (2005). The complex business of survival by aposematism, *Trends in Ecology & Evolution*, **20**, 598-603.

Martin, A. & Reed, R. D. (2014). Wnt signalling underlies evolution and development of the butterfly wing pattern symmetry systems, *Developmental Biology*, **395**, 367-378.

Mascalzoni, E., Osorio, D., Regolin, L. & Vallortigara, G. (2015). Symmetry perception by poultry chicks and its implications for three-dimensional object recognition, *Proceedings of the Royal Society B-Biological Sciences*, **279**, 841-846.

MATLAB and Statistics Toolbox Release 2012b, The MathWorks, Inc., Natick, Massachusetts, United States.

Mazo-Vargas, A., Concha, C., Livraghi, L., Massardo, D., Wallbank, R. W. R., Zhang, L., Papador, J. D., Martinez-Najera, D., Jiggins, C. D., Kronforst, M. R., Breuker, C. J., Reed, R. D., Patel, N. H., McMillan, W. O. & Martin, A. (2017). Macroevolutionary shifts of *WntA* function potentiate butterfly wing-pattern diversity, *Proceedings of the National Academy of Sciences of the United States of America*, **114**, 10701-10706.

Metzger, W. (1936). *Laws of Seeing*. *(English translation 2006)*. Cambridge, MA, MIT Press.

Merilaita, S. (1998). Crypsis through disruptive coloration in an isopod, *Proceedings of the Royal Society B Biological Sciences*, **265**, 1059-1064.

Merilaita, S. (2007). Artificial neural networks and the study of evolution of prey colouration, *Philosophical Transactions of the Royal Society B Biological Sciences*, **362**, 421-430.

Merilaita, S. & Lind, J. (2006). Great tits (*Parus major*) searching for artificial prey: implications for cryptic coloration and symmetry, *Behavioral Ecology*, **17**, 84-87.

Merilaita, S., Schaefer, H. M. Dimitrova, M. (2013). What is camouflage through distractive markings?, *Behavioral Ecology*, **24**, 1271-1272.

Merilaita, S., Scott-Samuel, N. E. & Cuthill, I. C. (2017). How camouflage works, *Philosophical Transactions of the Royal Society B*, **372**, 20160341.

Metzger, W. (1936). *Laws of Seeing. (English translation 2006).* Cambridge, MA, MIT Press.

Michalis, C. Scott-Samuel, N. E. Gibson, D. P. Cuthill, I. C. (2017). Optimal background matching camouflage, *Proceedings of the Royal Society B-Biological Sciences*, **284**, Article no. 20170709.

Møller, A. P. & Höglund, J. (1991). Patterns of fluctuating asymmetry in avian feather ornaments – Implications for models of sexual selection, *Proceedings of the Royal Society B-Biological Sciences*, **245**, 1-5.

Møller, A. P. (1992). Female swallow preference for symmetrical male sexual ornaments, *Nature*, **357**, 238-240.

Møller, A. P. (1993). Female preference for apparently symmetrical male sexual ornaments in the barn swallow *Hirundo rustica*, *Behavioral Ecology and Sociobiology*, **32**, 371-376.

Møller, A. P. & Pomiankowski, A. (1993). Fluctuating asymmetry and sexual selection, *Genetica*, **89**, 267-279.

Møller, A. P. & Eriksson, M. (1994). Patterns of fluctuating asymmetry in flowers – implication for sexual selection in plants, *Journal of Evolutionary Biology*, **7**, 97-113.

Møller, A. P. & Eriksson, M. (1995). Pollinator preference for symmetrical flowers and sexual selection in plants, *Oikos*, **73**, 15-22.

Monteiro, A., Brakefield, P. M. & French, V. (1997). The relationship between eyespot shape and wing shape in the butterfly *Bicyclus anynana*: a genetic and morphometrical approach, *Journal of Evolutionary Biology*, **10**, 787-802.

Neuwirth, E. (2014). RColorBrewer: ColorBrewer palettes. R package version 1.1-2. https://CRAN.R-project.org/package=RColorBrewer

Nijhout, H. F. (1980). Ontogeny of the color pattern on the wings of *Precis coenia* (Lepidoptera, Nymphalidae), *Developmental Biology*, **80**, 275-288.

Nokelainen, O. & Stevens, M. (2016). Camouflage. *Current Biology,* **26,** R654-R656.

Osorio, D. (1994). Symmetry versus crypsis, *Trends in Ecology & Evolution*, **9**, 346-346.

Osorio, D. (1996). Symmetry detection by categorization of spatial phase, a model, *Proceedings of the Royal Society B-Biological Sciences*, **263**, 105-110.

Osorio, D. & Cuthill, I. C. (2015). Camouflage and perceptual organization in the animal kingdom. *In:* Wagemans, J. (ed.) *The Oxford Handbook of Perceptual Organisation.* Oxford: Oxford University Press.

Palmer, A. R. & Strobeck, C. (1986). Fluctuating asymmetry – Measurement, analysis, patterns, *Annual Review of Ecology and Systematics*, **17**, 391-421.

Palmer, A.R. (1996). From symmetry to asymmetry: Phylogenetic patterns of asymmetry variation in animals and their evolutionary significance, *Proceedings of the National Academy of Sciences of the United States of America*, **93**, 14279-14286.

Paukner, A., Wooddell, L. J., Lefevre C. E., Lonsdorf, E. & Lonsdorf, E. (2017). Do capuchin monkeys (*Sapajus apella*) prefer symmetrical face shapes?, *Journal of Comparative Psychology*, **131,** 73-77.

Pietrewicz, A. T. & Kamil, A. C. (1977). Visual detection of cryptic prey by blue jays (*Cyanocitta cristata*), *Science*, **195**, 580-582.

Pietrewicz, A. T. & Kamil, A. C. (1979). Search image formation in the blue jay (*Cyanocitta cristata*), *Science*, **204**, 1332-1333.

Poulton, E. B. (1890). *The Colours of Animals: Their Meaning and Use. Especially Considered in the Case of Insects. Second Edition,* London, Kegan Paul, Trench Trübner, & Co. Ltd.

Prudic, K. L. Stoehr, A. M. Wasik, B. R. Monteiro, A. (2015). Eyespots deflect predator attack increasing fitness and promoting the evolution of phenotypic plasticity, *Proceedings of the Royal Society B-Biological Sciences,* **282**, Article no. 20141531.

R Core Team (2016). R: A language and environment for statistical computing. R Foundation for Statistical Computing, Vienna, Austria. URL https://www.R-project.org/.

Rainville, S. J. M. & Kingdom, F. A. (1999). Spatial-scale contribution to the detection of mirror symmetry in fractal noise, *Journal of the Optical Society of America A Optics Image Science and Vision*, **16**, 2112-2123.

Rainville, S. J. M. & Kingdom, F. A. A. (2000). The functional role of oriented spatial filters in the perception of mirror symmetry – psychophysics and modelling, *Vision Research*, **40**, 2621-2644.

Ramachandran, V. S., Tyler, C. W., Gregory, R. L., Rogers-Ramachandran, D., Duensing, S., Pillsbury, C. & Ramachandran, C. (1996). Rapid adaptive camouflage in tropical flounders, *Nature*, **379**, 815-818.

Rensch, B. (1958). Die Wirksamkeit ästhetischer Faktoren bei Wirbeltieren, *Zeitschrift für Tierpsychologie*, **14**, 71-00.

Rhodes, G. (2006). The evolutionary psychology of facial beauty, *Annual Review of Psychology*, **57**, 199-226.

Robertson, K. A. & Monteiro, A. (2005). Female *Bicyclus anynana* butterflies choose males on the basis of their dorsal UV-reflective eyespot pupils, *Proceedings of the Royal Society B-Biological Sciences*, **272**, 1541-1546.

Rock, I. (1983). *The Logic of Perception*, Cambridge: MIT.

Rodriguez, I., Gumbert. A., de Ibarra, N. H., Kunze, J. & Giurfa, M. (2004). Symmetry is in the eye of the 'beeholder': innate preference for bilateral symmetry in flower-naïve bumblebees, *Naturwissenschaften*, **91**, 374-377.

Rojas, B., Valkonen, J. & Nokelainen, O. (2015). Aposematism, *Current Biology*, **25**, 350-351.

Rosenholtz, R., Li, Y., Mansfield, J. & Jin, Z. (2005). Feature congestion: a measure of display clutter, In *Proceedings of the SIGCHI Conference on Human Factors in Computing Systems, ACM,* 761-770.

Rosenholtz, R., Li, Y. & Nakano, L. (2007). Measuring visual clutter, *Journal of Vision*, **7**, 1-22.

Ruxton, G. D., Allen, W. L., Sherratt, T. N. & Speed, M. P. (2018). *Avoiding Attack. 2nd edition.,* Oxford, Oxford University Press.

Ruxton, G. D., Sherratt, T. N. & Speed, M. P. (2004). *Avoiding Attack,* Oxford, Oxford University Press.

Sarkar, D. (2008). Lattice: Multivariate data visualization with R, Springer, New York. ISBN 978-0-387-75968-5.

Schaefer, H. M. & Stobbe, N. (2006). Disruptive coloration provides camouflage independent of background matching, *Proceedings of the Royal Society B-Biological Sciences,* **273,** 2427-2432.

Schluessel, V., Beil, O., Weber, Y. & Bleckmann, H. (2014). Symmetry perception in bamboo sharks (*Chiloscyllium griseum*) and Malawi cichlids (*Pseudotropheus sp.*), *Animal Cognition*, **17**, 1187-1205.

Schreiber, A. M. (2013). Flatfish: An asymmetric perspective on metamorphosis, *Animal Metamorphosis*, **103**, 167-194.

Shohet, A. J., Baddeley, R. J., Anderson, J. C. & Osorio, D. (2007). Cuttlefish camouflage: a quantitative study of patterning, *Biological Journal of the Linnean Society*, **92**, 335-345.

Shohet, A. J. Baddeley, R. J., Anderson, J. C., Kelman, E. J. & Osorio, D. (2006). Cuttlefish responses to visual orientation of substances, water flow and model of motion camouflage, *Journal of Experimental Biology,* **209**, 4717-4723.

Steiner, C. C., Weber, J. N. & Hoekstra, H. E. (2007). Adaptive variation in beach mice produced by two interacting pigmentation genes, *PLOS Biology*, **5**, 1880-1889.

Sterry, P., Cleave, A. & Read, R. (2016). *British Butterflies & Moths (Collins Complete Guides)*, London: William Collins 2016.

Stevens, M. (2016). *Cheats and Deceits*, Oxford, Oxford University Press.

Stevens, M., Castor-Perry, S. A. & Price, J. R. F. (2009). The protective value of conspicuous signals is not impaired by shape, size, or position asymmetry, *Behavioral Ecology*, **20**, 96-102.

Stevens, M., Cuthill, I. C., Windsor, A. M. M. & Walker, H. J. (2006). Disruptive contrast in animal camouflage, *Proceedings of the Royal Society B-Biological Sciences,* **273**, 2433-2438.

Stevens, M., Graham, J., Winney, I. S. & Cantor, A. (2008). Testing Thayer's hypothesis: can camouflage work by distraction?, *Biology Letters*, **4**, 648-650.

Stevens, M., Marshall, K. L.A., Troscianko, J., Finlay, S., Burnand, D. & Chadwick, S. L. (2013a). Revealed by conspicuousness: distractive markings reduce camouflage, *Behavioral Ecology*, **24**, 213-222.

Stevens, M., Troscianko, J., Marshall, K. L. A. & Finlay, S. (2013b). What is camouflage through distractive markings? A reply to Merilaita et al. (2013), *Behavioral Ecology*, **24**, 1272-1273.

Stevens, M. & Merilaita, S. (2009a). Animal camouflage: current issues and new perspectives, *Philosophical Transactions of the Royal Society B Biological Sciences*, **364**, 423-427.

Stevens, M. Merilaita, S. (2009b). Defining disruptive coloration and distinguishing its functions, *Philosophical Transactions of the Royal Society B-Biological Sciences,* **364,** 481-488.

Stevens, M. Merilaita, S. (2011). *Animal Camouflage: Mechanisms and Function.,* Cambridge, UK, Cambridge University Press.

Stevens, M., Parraga, C. A., Cuthill, I. C., Partridge, J. C. & Troscianko, T. S. (2007). Using digital photography to study animal coloration, *Biological Journal of the Linnean Society*, **90**, 211-237.

Swaddle, J. P. & Cuthill, I. C. (1994a). Preference for symmetrical males by female zebra finches, *Nature*, **367**, 165-166.

Swaddle, J. P. & Cuthill, I. C. (1994b). Female zebra finches prefer males with symmetrically manipulated chest plumage. *Proceedings of the Royal Society B-Biological Sciences*, **258**, 267-271.

Swaddle, J. P. (1999). Visual signalling by asymmetry: a review of perceptual processes, *Philosophical Transactions of the Royal Society of London B-Biological Sciences*, **354**, 1383-1393.

Swaddle, J. P. & Pruett-Jones, S. (2001). Starlings can categorize symmetry differences in dot displays, *American Naturalist*, **158**, 300-307.

Swaddle, J. P. & Ruff, D. A. (2004). Starlings have difficulty in detecting dot symmetry: Implications for studying fluctuating asymmetry, *Behaviour*, **141**, 29-40.

Swaddle, J. P., Ruff, D. A., Page, L. C., Frame, A. M. & Long, V. A. (2008). A test of receiver perceptual performance: European starlings' ability to detect asymmetry in a naturalistic trait, *Animal Behaviour*, **76**, 487-495.

Sweeney, A., Jiggins, C. & Johnsen, S. (2003). Insect communication: Polarized light as a butterfly mating signal, *Nature*, **423**, 31-32.

Tagawa, M. & Aritaki, M. (2005). Production of symmetrical flatfish by controlling the timing of thyroid hormone treatment in spotted halibut *Verasper variegatus*, *General and Comparative Endocrinology*, **141**, 184-189.

Terry, M., Therneau, T. & Patricia, M. G. (2000). Modelling survival data: Extending the Cox model, *Springer. New York*. ISBN 0-387-98784-3.

Thayer, G. H. (1909). *Concealing-coloration in the animal kingdom: an exposition of the laws of disguise through color and pattern: being a summary of Abbott H. Thayer's discoveries.,* New York, Macmillan.

Therneau, T. (2015). A package for survival analysis in S version 2.38, URL: https://CRAN.R-project.org/package=survival

Therneau, T. M. (2018). coxme: Mixed effects Cox models. R package version 2.2-7. See: https://CRAN.R-project.org/package=coxme

Troscianko, T., Benton, C. P., Lovell, P. G., Tolhurst, D. J. & Pizlo, Z. (2009). Camouflage and visual perception, *Philosophical Transactions of the Royal Society B-Biological Sciences*, **364**, 449-461.

Troscianko, J. & Stevens, M. (2015). Image calibration and analysis toolbox – a free software suite for objectively measuring reflectance, colour and pattern, *Methods in Ecology and Evolution*, **6**, 1320-1331.

Tyler, C. W., Hardage, L. & Miller, R. T. (1995). Multiple mechanisms for the detection of mirror symmetry, *Spatial Vision*, **9**, 79-100.

Umbers, K. D. L., Lehtonen, J. & Mappes, J. (2015). Deimatic displays, *Current Biology*, **25**, 58-59.

Von Fersen, L., Manos, C. S., Goldowsky, B. & Roitblat, H. (1992). Dolphin detection and conceptualization of symmetry, in *Marine Mammal Sensory Systems*, 753-762, (ed. Thomas, J. *et al.*) (Plenum, New York 1992).

Wallace, A. R. (1867). Mimicry, and other protective resemblances among animals, *Westminster Reviews*, **32**, 1-43.

Wallace, A. R. (1877). The colors of animals and plants. Part I. *The American Naturalist,* **11,** 384-406.

Wallace, A. R. (1879). The protective colours of animals. In *Science for all*, ed. Robert Brown, **2**, 284-296. London: Cassel, Petter, Galpin.

Wallace, A. R. (1889). *Darwinism. An Exposition of the Theory of Natural Selection With Some of its Applications*, London, Macmillan & Co.

Wallace, A. R. (1891). Natural selection and tropical nature, essays on descriptive and theoretical biology, London and New York: Macmillan.

Webster, R. J., Callahan, A., Godin, J. G. J. & Sherratt, T. N. (2009). Behaviourally mediated crypsis in two nocturnal moths with contrasting appearance, *Philosophical Transactions of the Royal Society B-Biological Sciences,* **364**, 503-510.

Wenderoth, P. (1994). The salience of vertical symmetry, *Perception*, **23**, 221-236.

Wilkinson, F., Wilson, H. R. & Habak, C. (1998). Detection and recognition of radial frequency patterns, *Vision Research*, **38**, 3555-3568.

Wilson, H. R. & Wilkinson, F. (2002). Symmetry perception: a novel approach for biological shapes, *Vision Research*, **42**, 589-597.

Windig, J. J. & Nylin, S. (1999). Adaptive wing asymmetry in males of the speckled wood butterfly (*Pararge aegeria*)? *Proceedings of the Royal Society B-Biological Sciences*, **266**, 1413-1418.

Wittkopp, P. J. & Beldade, P. (2008). Development and evolution of insect pigmentation: Genetic mechanisms and the potential consequences of pleiotropy, *Seminars in Cell & Developmental Biology*, **20**, 65-71.

Wright, S. (1932). The roles of mutation, inbreeding, crossbreeding and selection in evolution, *Proceedings of the Sixth International Congress of Genetics,* **1**, 356-366.

Xiao, F. & Cuthill, I. C. (2016). Background complexity and the detectability of camouflaged targets by birds and humans, *Proceedings of the Royal Society B-Biological Sciences*, **283**, Article no. 20161527.

Zhang, S., Mao, K. Lin, P., Ho, C., Hung, W., Piorkowski, D., Liao, C. & Tso, I. (2015). Crypsis via leg clustering: twig masquerading in a spider, *Royal Society Open Science,* **2**, 150007.

Zylinski, S., Darmaillacq, A. S. & Shashar, N. (2012). Visual interpolation of contour completion by the European cuttlefish (*Sepia officinalis*) and its use in dynamic camouflage, *Proceedings of the Royal Society B-Biological Sciences*, **279**, 2386-2390.

Zylinski, S., Osorio, D. & Johnsen, S. (2016). Cuttlefish see shape from shading, fine-tuning coloration in response to pictorial depth cues and directional illumination, *Proceedings of the Royal Society B-Biological Sciences*, **283**, Article no. 20160062.

Zylinski, S., Shohet, A. & Osorio, D. (2009). The use of multiple visual cues in cuttlefish camouflage, *Perception*, **38**, 89.